## Esthetic Management of Incisors with Diffuse and Demarcated Opacities: 24 Month Follow-up Case Report

GS Athayde • RC Jorge • GCA Americano • F Barja-Fidalgo • VM Soviero

#### Clinical Relevance

A minimally invasive esthetic treatment can provide longevity and color stability.

#### **SUMMARY**

#### This clinical case report describes a minimally invasive approach to mask diffuse and demar-

Glaucia dos Santos Athayde, DDS, MSc, PhD student, Department of Preventive and Community Dentistry, Dental School, Universidade do Estado do Rio de Janeiro UERJ & UNIFESO, Rio de Janeiro, Brazil

Roberta Costa Jorge, DDS, MSc, PhD student, Department of Preventive and Community Dentistry, Dental School, Universidade do Estado do Rio de Janeiro UERJ, Faculdade Arthur Sá Earp Neto, Rio de Janeiro, Brazil

Gabriela Caldeira Andrade Americano, DDS, MSc, PhD student, Department of Preventive and Community Dentistry, Dental School, Universidade do Estado do Rio de Janeiro UERJ & UNILESTE, Rio de Janeiro, Brazil

Fernanda Barja-Fidalgo, DDS, MSc, PhD, professor, Department of Preventive and Community Dentistry, Dental School, Universidade do Estado do Rio de Janeiro UERJ, Rio de Janeiro, Brazil

\*Vera Mendes Soviero, DDS, MSc, PhD, professor, Department of Preventive and Community Dentistry, Dental School, Universidade do Estado do Rio de Janeiro UERJ & Faculdade Arthur Sá Earp Neto, Rio de Janeiro, Brazil

\*Corresponding author: Av 28 deSetembro 157, 2° andar - Vila Isabel, Rio de Janeiro 20551-030, Brazil; e-mail: verasoviero@gmail.com

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cated opacities in permanent anterior teeth in an eight-year-old child who was upset with the appearance of her incisors. Clinical examination showed diffuse opacities in teeth 7, 8, 9, and 10, diagnosed as dental fluorosis associated with yellow demarcated opacity in tooth 9 related to molar-incisor hypomineralization. The treatment was based on conservative dentistry using the low-viscosity resin infiltration technique and resin composite restoration in an attempt to mask the lesions. The followup time was 24 months.

#### INTRODUCTION

Developmental defects of enamel in anterior teeth might have negative impact on patient self-esteem and consequently quality of life. 1,2 Developmental defects of enamel can affect all the teeth as a whole or can be localized in a specific group of teeth, as in molar incisor hypomineralization (MIH). MIH is characterized by the presence of demarcated opacities in at least one first permanent molar, generally associated with incisors also affected. Fluorosis presents diffuse opacities with a chalky white appearance or, in severe cases, with staining and pitting. A demarcated opacity in a single permanent tooth can be attributed to trauma to its predeces-

sor.<sup>3,7</sup> All these conditions come from abnormality in the enamel mineralization phase, modifying the chemical composition and optical characteristics of the enamel.<sup>7</sup>

The patient's expectations and the severity of the enamel opacities will guide the clinician's treatment choice. Alternatives to achieve esthetic improvement of demarcated and diffuse opacities range from minimally invasive strategies, such as dental bleaching<sup>8,9</sup> and microabrasion, <sup>9-11</sup> to more invasive techniques, such as composite resin restorations or porcelain veneers. 12,13 Microabrasion is indicated for fluorotic opacities; however, care should be taken to avoid excessive removal of enamel during the clinical procedure. 13 Among the minimally invasive methods, bleaching with hydrogen peroxide or carbamide peroxide has been used successfully. 14 Composite resins and porcelain veneers have been used to provide good esthetic results but invariably cause greater loss of tooth structure due to tooth preparation. 12,13

More recently, the use of an infiltrant resin has been recommended as a minimally invasive approach to masking opacities in the enamel. The infiltrant agent was initially proposed for the treatment of white spot lesions caused by dental caries, but successful experiments have been reported for masking opacities related to fluorosis and opacities caused by trauma. The technique of infiltrating the opacity with low-viscosity resin is based on the fact that the resin has a refractive index similar to that of sound enamel. Thus, when the resin penetrates and fills the porosities of the carious lesions, the refractive index returns to values very close to those of the sound enamel, masking the opacity. <sup>18</sup>

This clinical case report describes a minimally invasive approach to mask diffuse and demarcated opacities in permanent anterior teeth using a resin infiltrant (Icon, DMG, Hamburg, Germany) and composite resin restoration.

#### **CLINICAL CASE REPORT**

An eight-year-old female patient with no systemic disease attended the Pediatric Dentistry Clinic of Rio de Janeiro State University, Rio de Janeiro, Brazil. She was upset with the whitish appearance of her maxillary permanent incisors. Clinical examination showed diffuse opacities in teeth 7, 8, 9, and 10, which was diagnosed as moderate fluorosis (score 5) based on the Thylstrup and Fejerskov Index for Dental Fluorosis. <sup>19</sup> A yellow demarcated opacity in



Figure 1. Baseline: diffuse opacities in the upper central and lateral incisors. Yellow demarcated opacity in tooth 9.

tooth 9 was diagnosed as MIH, according to the European Academy of Pediatric Dentistry<sup>4</sup> because the first permanent molars were also affected (Figure 1).

The treatment decision was based on minimally invasive dentistry, using the infiltration technique with low-viscosity resin (Icon) and composite resin (TPH, Dentsply, São Paulo, Brazil) in an attempt to mask the discolorations. The parent signed an informed consent form authorizing the treatment and the use of images.

The teeth were cleaned and isolated with a rubber dam (Figure 2). The infiltration technique followed was per the manufacturer's instructions. The enamel was etched using 15% HCl gel (Icon etch, DMG) for 2 minutes (Figure 3) followed by rinsing for 30 seconds, drying with compressed air (Figure 4) and dehydration with 100% ethanol (Icon dry, DMG) for 30 seconds. During ethanol application the visual aspect of the enamel was checked and the opacities had not disappeared. Thus, the etching step was repeated. After the second acid etching, the diffuse opacities were masked, but the MIH yellow opacity in tooth 9 was still visible. Then, approximately 0.2 mm of the enamel was removed with a diamond bur and the acid gel (Icon etch) was reapplied only in the area of the demarcated opacity (Figures 5 and 6). When ethanol was applied, it was possible to see that the demarcated opacity was also camouflaged (Figure 7). The infiltration resin (Icon Infiltrant, DMG) was carefully applied onto the etched area for 3 minutes without rubbing (Figure 8); slightly dried with compressed air; and, after removing the excess with gauze, light-cured for 40 seconds (Altlux, Ribeirão Preto, São Paulo, Brazil) (Figure 9). Composite resin, color A2, (TPH) was used to restore the enamel removed by the bur. The tooth surfaces were polished with composite resin polishing discs (Sof-Lex, 3M ESPE, St Paul, MN, USA). The clinical

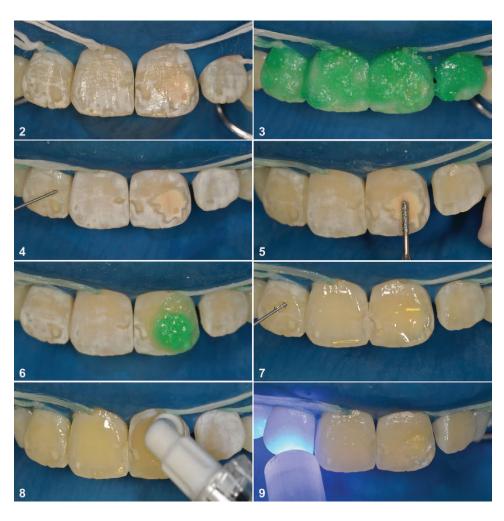


Figure 2. Rubber dam with ligatures.

Figure 3. 15% hydrochloric acid application (2 min).

Figure 4. Aspect after hydrochloric acid application. Ready for ethanol application.

Figure 5. After ethanol application, the diffuse white areas disappeared. The demarcated opacity did not change the aspect and a thin layer of enamel was removed (diamond bur-tooth 9).

Figure 6. 15% hydrochloric acid application in the demarcated opacity – tooth 9 for 2 min.

Figure 7. Ethanol application to preview the masking effect.

Figure 8. Resin infiltration in teeth 7, 8, 9, and 10.

Figure 9. Light-cure for 40s.

aspect immediately after infiltration and rubber dam removal and at the 12-month and 24-month followup are shown in Figures 10, 11, 12, and 13 respectively.

#### **DISCUSSION**

Resin infiltration is an approach that can be safely and effectively performed in a single session with optimal results.  $^{15\text{-}17,20}$  The diffuse opacities were completely masked by the infiltrant. Compared with microabrasion, the resin infiltration technique removes considerably less enamel as the acid application is not combined with mechanical abrasion. The microabrasion technique with abrasive gel composed of a mixture of hydrochloric acid or phosphoric acid and pumice or silica carbide particles is widely used as a minimal intervention,  $^{21,22}$  although it invariably results in considerable enamel reduction. The enamel loss after microabrasion ranges from around  $100~\mu m$  to more than  $250~\mu m$  depending on the pressure, time of application, and concentration of the acid.  $^{21,23\text{-}26}$  As mild to moderate fluorosis

presents a porous subsurface zone of 80 to 100 µm in depth, microabrasion abrades the whole affected enamel. Hence, the esthetic appearance is improved because the fluorotic enamel is removed. Etching with 15% HCl for 120 seconds, according to the resin infiltration technique, removes between 30 and 40 um of surface layer, thus enabling the resin to penetrate into the deeper affected enamel.<sup>27</sup> The masking effect occurs due to the resin infiltrating the porous subsurface. That is why the application of ethanol is used as a prognostic parameter of the final result. If the wetting effect of the ethanol masks the opacities partially or completely, it means that the infiltrant will penetrate and mask the opacities. For a thicker surface zone additional etching may be necessary, as in the present case.

Home bleaching has also been recommended as an esthetic treatment for fluorosis. The modality that involves the use of a vacuum-formed custom-fitted tray filled with carbamide peroxide (10%-20%) or hydrogen peroxide (1%-10%) and has been validated by several studies. <sup>14,28,29</sup> For pediatric patients, the



Figure 10. Immediate aspect after resin infiltration and composite resin – teeth 7, 8, 9 and 10.

Figure 11. Immediate aspect after rubber dam removal.

Figure 12. 6-month follow-up.

Figure 13. 24-month follow-up.

American Association of Pediatric Dentistry recommends caution in the use of bleaching techniques.<sup>30</sup> Taking into account that in young patients dental pulp is wider than in adult patients and that the diffusion of hydrogen peroxide at high concentrations into dental tissues is extensive, the risk of adverse effects such as sensitivity might be high. Additionally, in contrast to bleaching therapy, which can reduce the microhardness of demineralized enamel surfaces,<sup>31</sup> the infiltrant resin can strengthen the enamel structure mechanically.<sup>32</sup> Therefore, for the current patient, bleaching was not considered.

In contrast to the fluorotic opacities, the demarcated opacity localized in the left incisor did not show any improvement even after a second acid etching. It was decided to use a bur to remove the superficial layer of the opacity and repeat the acid etching. In this case, treatment combining resin infiltration with composite can be considered a minimally invasive procedure because a very thin layer of enamel was removed that was restricted to the area of the demarcated opacity. Microabrasion was not used because of concerns about the risk of postoperative sensitivity considering the amount of enamel that would need to be removed and the patient's young age. However, in a previous case report, a nine-year-old patient had a demarcated opacity that was treated with microabrasion repeated in three consecutive appointments followed by resin infiltration; the patient had no complaints about sensitivity.33

The combination of resin infiltration and composite was effective in improving esthetics with minimal loss of tooth structure. Once the demarcated opacity was masked by the infiltrant, the restoration could be done with a single-color composite in a simple and

practical procedure. No strategies combining opaque liner and different colors of composite were necessary to obtain the masking effect. Additionally, as bonding on a surface previously treated with Icon is possible,<sup>34</sup> no primer or adhesive was used before inserting the composite resin. The esthetic result showed color stability at the 24-month follow-up.

MIH is relatively common worldwide with an average prevalence of 11.24%,<sup>35</sup> which means pediatric dentists and clinicians will probably have patients with esthetic issues related to this condition and should be aware of possible treatment options. Considering the importance of the smile during adolescence and implications for self-esteem and social well-being to a child, clinicians should consider a conservative and effective approach. In the current case, diffuse and demarcated opacities were masked by the combination of infiltrant with composite in a single session, and a satisfactory outcome was achieved.

#### **CONCLUSIONS**

The esthetic treatment based on resin infiltration and composite achieved excellent results with color stability at the 24-month follow-up. Diffuse and demarcated opacities were masked in a single session that improved the patient's self-esteem.

#### Acknowledgement

Special thanks to DMG (Hamburg, Germany) for providing Icon.

#### **Regulatory Statement**

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of the ethical committee from Universidade do Estado do Rio de Janeiro. The approval code issued for this study is 07753213.0.0000.5259.

#### **Conflict of Interest**

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## A Simple Method for Modifying the Emergence Profile by Direct Restorations: The Biologically Active Intrasulcular Restoration Technique

L Giachetti

#### **Clinical Relevance**

It is possible to modify the natural emergence profile of the tooth using simple intrasulcular direct restorations. The shape of the intrasulcular part of the restoration will determine the design of the gingival contour.

#### **SUMMARY**

Some clinical situations, such as the closure of pronounced diastemas and the transformation of malformed, small, or peg-shaped teeth, require a rebalancing of dental proportions accompanied by a modification of the gingival contour. A traditional treatment plan might require surgical, prosthetic, and/or orthodontic treatment, but in some cases, these therapeutic options could be considered too invasive and not always the best solution. Moreover, not all patients are ready to undergo irreversible, long, and expensive procedures. To overcome these limitations and to

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#### INTRODUCTION

Generally, when it comes to applying adhesive techniques, both direct and indirect, one of the requirements is that the margin of the restoration is in an extrasulcular position. This leads to a number of advantages such as improved moisture

solve all of these clinical problems in a rapid and noninvasive way, we propose a new technique that allows us to modify the natural emergence profile of the tooth using simple intrasulcular direct restorations. Using the Biologically Active Intrasulcular Restoration technique, it is possible to rebalance tooth shape and dimensions, gingival level and contour with low biological and economic costs. This method, which does not require any preparation of the dental tissues, is reversible and minimally invasive. It is applicable to patients of all ages, and results are obtained in a single appointment.

<sup>\*</sup>Luca Giachetti, MD, DMD, MSc, PhD, associate professor, Dental Materials and Restorative Dentistry, University of Florence Department of Experimental and Clinical Medicine -Unit of Dentistry, Firenze, Italy

<sup>\*</sup>Corresponding author: Via del Ponte di Mezzo 48 - 50127 Firenze, Italy; e-mail: luca.giachetti@unifi.it

control (easier application of the rubber dam) and respect for periodontal health.

However, there are some clinical situations in which, to achieve satisfying esthetic results, it would be necessary to put the restoration margin apically with respect to the gingival line. This condition occurs in several clinical situations, such as the closure of pronounced diastemas or the transformation of malformed, small, or peg-shaped teeth, clinical situations in which a rebalance of dental proportions is required, very often accompanied by a modification of the gingival contour.

A traditional treatment plan might require surgical, prosthetic, and/or orthodontic treatment, but in some cases, these therapeutic options could be considered too invasive and not always the best solution. Moreover, not all patients are in a position to undergo irreversible, long, and expensive procedures.

To overcome these limitations and to solve these clinical problems in a rapid and noninvasive way, we propose a new technique that allows us to modify the natural emergence profile of the tooth using simple intrasulcular direct restorations: The Biologically Active Intrasulcular Restoration (BAIR) technique. The BAIR technique allows us to perform restorations that can actively interact with the surrounding soft tissues and facilitate the adaptation of the gingival margins. By modifying the emergence profile we are able to change the position of the gingival margin since the restoration holds the surrounding soft tissues. The shape of the intrasulcular part of the restoration will determine the design of the gingival contour.

This procedure enables us to apically displace the soft tissues, thus resulting in a lengthening of the clinical crown and an immediate reshaping of the gingival contour, helping to restore more appropriate dental proportions.

#### **Purpose**

The aim of this article is to describe the BAIR technique and to show its clinical applications, together with its advantages.

#### **Description of Technique**

The proposed method makes use of a circular metal matrix to isolate the operative site and, at the same time, to move the soft tissues and provide clear access to the intrasulcular portion of the tooth. By doing so, a proper isolation is obtained, allowing the application of adhesives and composites.

#### CASE 1

A young girl showed poor alignment of the two central incisors. The left central incisor seemed to be extruded with respect to the adjacent teeth. The most appropriate therapy could be orthodontic treatment, which the patient had no intention of undertaking. Anamnesis told us that this tooth had a fracture and a subsequent reattachment of the fragment. At the time, there was evident discoloration, with more opaque and high chromatic dentin (Figure 1A). The tooth had been shortened, and space had been created to achieve more translucency. The metal matrix was applied, and the gingiva was pushed apically. The matrix was positioned around the tooth, tilted, pushed in the cervical direction keeping it adjacent to the tooth, and slid up to fit into the gingival sulcus. The matrix adaptation was facilitated with the use of an excavator or tweezers (Figure 1B).

Once in the sulcus, the metal matrix was gently pushed again to move the soft tissues. In this way, it was possible to obtain an isolation of the operative site and dislocation of the soft tissues in the apical, mesial, and distal directions. This process provided clear access to the intrasulcular portion of the tooth (Figure 1C) and allowed easy application of adhesive and composite. These applications facilitated the rebuilding of a new artificial cementoenamel junction (CEJ), which changed the emergence angle between the root and crown (Figure 1D). To avoid overhangs, the first layer had to be applied while the edge of the matrix was in close contact with the tooth.

The junction between the root and composite was moved beyond the natural CEJ. A new hybrid CEJ was re-created more apically, so that the gingiva could be supported and adapt itself to the design that had been created with the composites (Figure 1E). Just after removing the matrix, it was noted that the incisal and gingival levels were reestablished. The finishing of the extragingival part of the restoration was performed, whereas the intrasulcular part did not need finishing or polishing as it was already smooth and well cured because the composite polymerized in contact with the metal matrix and in the absence of oxygen (Figure 1F). After 2 years, the patient's gingival health was stable. Unfortunately, the discoloration was not properly improved. In hindsight, a greater amount of dentin should have been removed to reduce the opacity of the tooth (Figure 1G).

#### CASE 2

A young patient who was at the end of orthodontic treatment needed and wished for an enlargement of

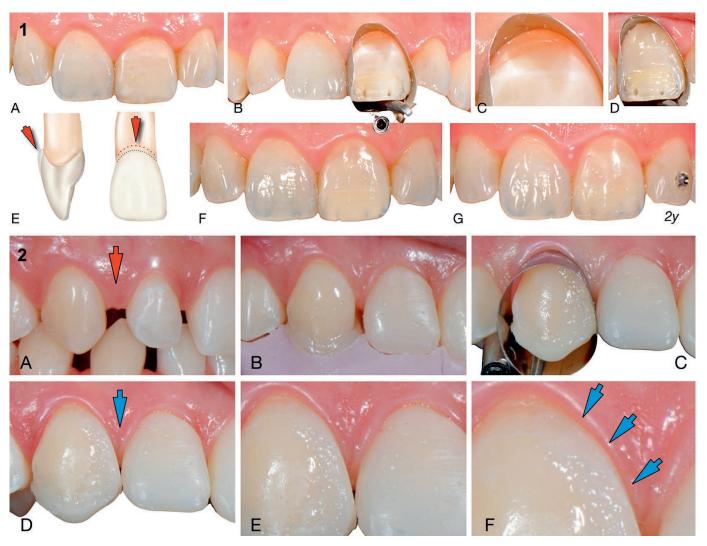


Figure 1. (A): Poor alignment of the two central incisors. The left central incisor seems to be extruded with respect to the adjacent teeth. (B): The tooth has been shortened, and space has been created to obtain more translucency. The metal matrix is applied, and the gingiva is pushed apically. (C): The cervical area is isolated. (D): The composite is applied and pressed against the matrix and the tooth to change the emergence profile between the root and the crown. (E): In the diagram, it is highlighted that the border between the root and composite has been moved apically (red arrows). A new "hybrid cementoenamel junction" is created more apically, so that the gingiva can be supported and can adapt itself to the design that has been created with composites (red dotted line). (F): Just after removing the matrix, it can be noted that the incisal and gingival levels were reestablished. (G): After 2 years, the patient has stable gingival health.

Figure 2. (A): A very evident diastema between the canine and lateral incisor (red arrow). (B): Using a template, prepared through a wax-up, we can rebuild the incisal edge with the new mesiodistal dimension. (C): The restoration was completed by connecting the incisal part with the cervical one, with the help of the metal matrix. (D): In this way, the proximal walls emerge from within the gingival sulcus. The diastema is closed (blue arrow). (E, F): It can be noted how the composite supports the gingival tissue (blue arrows) and how in this way we can instantly obtain a papilla.

her tiny teeth (Figure 2A). A wax-up was carried out, and from this, a template was prepared. This helped us to realize the incisal edge with the new mesiodistal dimension (Figure 2B). The restoration was completed by connecting the incisal part to the cervical one, with the help of a metal matrix (Figure 2C). In this way, the proximal walls could emerge from within the gingival sulcus (Figure 2D). It can be noted that the composite supported the gingival

tissue, and in this way, the papilla was instantly obtained (Figure 2E,F).

#### CASE 3

The same procedure is applicable when the restoration of gingival papilla is desired because of the absence of an interproximal contact point. An elderly patient, wearing a removable prosthesis, showed an unpleasant smile because of small and worn incisors



Figure 3. (A): Unpleasant smile due to small and worn incisors and absence of papillae. (B): In a single appointment, using the Biologically Active Intrasulcular Restoration technique, the shape of the teeth was changed, thus creating new contacts more apically with consequent better adaptation of the soft tissues.

and the absence of papillae. Her poor economic resources did not allow her to pay for a complex treatment plan (Figure 3A). Using the BAIR technique, the shape of her teeth was changed, thus creating new contacts more apically with consequent better adaptation of the soft tissues (Figure 3B). Her esthetic problems were solved in a single appointment.

#### CASE 4

A young patient with a peg-shaped upper left lateral incisor was treated using the BAIR technique (Figure 4A). The metal matrix allowed us to obtain insulation of the operative site and, at the same time, dislocation of the soft tissues in the apical, mesial, and distal directions (Figure 4B,C). Excess composite was applied because of the position and shape of the matrix (Figure 4D). The cervical part of the restoration drew the gingival contour and kept the surrounding soft tissues in a more apical position (Figure 4E). The right volumes and shape were restored afterward using a subtractive modeling technique, without milling the intrasulcular part of the restoration (Figure 4F). Changing the emergence angle of the tooth, together with its shape and contact points, allowed us to obtain a rapid adaptation of the soft tissue, with the restoration itself drawing the new gingival contour (Figure 4G).

Follow-up after 2 years showed stable gingival health (Figure 4H-J).

#### **MATERIALS USED**

We used the Automatrix Medium Regular Band (Dentsply/Caulk, Milford, DE, USA). The adhesive system was chosen depending on the substrate. If only enamel needed to be conditioned, bonding resin (Heliobond, Ivoclar Vivadent, Schaan, Liechtenstein) was applied after conditioning with orthophosphoric acid (Gel Etchant, Kerr, Orange, CA, USA) for 30 seconds. If the restoration margin was located beyond the CEJ on a dentin substrate, a selective etching on enamel was performed, and a self-etch adhesive containing 10-MDP was applied (Clearfil SE, Kuraray, Hattersheim am Main, Germany). A medium-opacity universal composite was used (Supreme XTE, 3M ESPE, St Paul, MN, USA). Once the matrix was removed, a subtractive modeling was performed with a fine diamond bur (Composhape 4236 40 µm, Intensiv SA, Lugano, Switzerland). Finishing was performed with abrasive disks (Sof-Lex Pop-On XT 2381 M Ø 9.5mm, 3M ESPE) and abrasive strips (Proxostrip 40-15, Intensiv SA, Lugano, Switzerland). The restoration was then polished with a polishing paste (Optra-Fine HP, Ivoclar Vivadent) using a round brush (Enamel Plus Shiny S, Micerium, Avegno, Genova, Italy).

#### POTENTIAL PROBLEMS

The use of a traditional dental dam is not indicated because it is likely to hinder the vision of the gingival parabolas of the teeth, while it is essential to be able to continuously check the soft tissues to achieve a correct balancing of the gingival levels. However, the localized isolation offered by the metal matrix is more than sufficient, and in the BAIR technique, the restoration originates in an area that would not be isolated by the dam anyway.

Generally, the metal matrix was stabilized with wooden wedges or by simply taking advantage of the contact points of the adjacent teeth. However, sometimes there were no adjacent teeth or we needed to significantly modify the position of the soft tissues. In such cases, it was necessary for the operator to exert a constant pressure on the matrix, using a finger to keep it in the desired position until the first composite layer was light cured. Therefore, constant assistance needed to be available.

If it was necessary to complete the restoration of the palatal side of the interproximal walls, we used

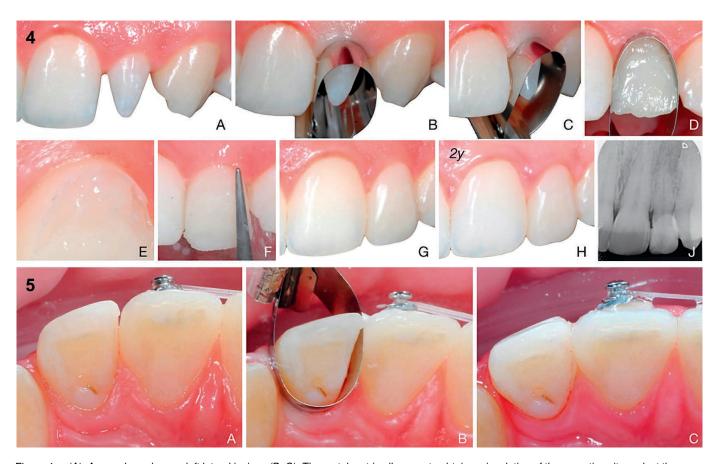


Figure 4. (A): A peg-shaped upper left lateral incisor. (B, C): The metal matrix allows us to obtain an insulation of the operative site and, at the same time, the dislocation of the soft tissues in the apical, mesial, and distal directions. Gingival ischemia can be noted in the compression zone, which occasionally remains for a short period after the removal of the matrix. Within a few hours, the gingiva regains a healthy and natural appearance. (D): Excess composite is applied because of the position and shape of the matrix. (E): The matrix has just been removed. At high magnification, it is evident how the restoration supports the surrounding soft tissues. (F): The right volumes and shape are restored using a subtractive modeling technique, without milling the intrasulcular part of the restoration. (G): The final result. The restoration draws the new gingival contour. (H): After 2 years, stable gingival health is maintained. (J): X-ray at 2 years.

Figure 5. (A): After completing the closure of a diastema on the vestibular side, a defect remains on the mesiopalatal side. (B): The matrix was inclined toward the sulcus from the lingual side to isolate the defect and allow the application of the composite. (C): The defect was repaired.

the matrix inclined toward the sulcus from the lingual side (Figure 5A-C).

Another issue could be the periodontal tissue adaptation to the intrasulcular restorations. Scientific evidence suggests that if such a restoration is carried out through careful control of local humidity; superficial polymerization, polishing and finishing; and local hygiene, then it can perfectly integrate with the surrounding periodontium, without being itself a cause of inflammation.<sup>2-9</sup> In the BAIR technique, the circular metal matrix, together with other isolation disposables, provides local moisture control and allows an adequate curing of the restorative materials. Moreover, the contact between the matrix and the composite ensures the realization of a perfectly smooth area in the most apical part of the restoration, which does not require any finishing

and polishing. In our clinical experience, none of the treated patients revealed periodontal problems in the site of the restorations.

### SUMMARY OF ADVANTAGES AND DISADVANTAGES

#### **Advantages**

The use of a metal matrix provides valid local isolation, displaces the gingiva in an atraumatic way, and allows clear access to the intrasulcular portion of the tooth.

The subgingival portion of the restoration is perfectly smooth because it is polymerized in contact with the metal matrix, in the absence of oxygen. For this reason, the intrasulcular part of the restoration does not need finishing or polishing, avoiding

superficial roughness that may interfere with the adaptation of periodontal tissues to the restoration.  $^{10-15}$ 

No surgery is required, and more often than not, anesthesia is not needed.

Since there is no reduction of the dental tissues, this technique is minimally invasive and, if desired, allows us to restore the initial situation.

It is possible to easily modify the restorations later, if necessary, using the same protocol as for the first intervention.

#### **Disadvantages**

When there is a need to modify heavily the position of the soft tissues, it is necessary for the operator to exert a constant pressure on the matrix by a finger to keep the matrix in the desired position. This means having constant assistance available.

This technique is indicated for single-rooted teeth only, and it cannot be applied to teeth that are part of a splinting.

#### CONCLUSION

With simple direct restorations, we can achieve results that would otherwise (ie, with traditional treatment plans) require surgery or orthodontics and much longer treatments often associated with annoying postoperative problems.

Using the BAIR technique, we can rebalance tooth shape and dimensions as well as gingival level and contour with low biological and economic costs. The method does not require any preparation of the dental tissues. It is reversible and minimally invasive. It is applicable to patients of all ages, and results are obtained in a single appointment.

#### **Conflict of Interest**

The author of this article certifies that he has no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Temporary Tooth Separation to Improve Assessment of Approximal Caries Lesions: A School-Based Study

MM Nascimento • AP Ribeiro • AJ Delgado • L Cassiano • MGD Caraballo J-F Roulet • S Geraldeli • PNR Pereira • ME Ottenga • DA Dilbone

#### Clinical Relevance

Radiographic lesion depth should not be used as the single determinant of the restorative threshold for clinically inaccessible approximal caries lesions. Temporary tooth separation is a feasible and effective diagnostic aid for assessment and appropriate management of approximal lesions.

#### **SUMMARY**

In the era of tooth-preserving dentistry, the decision to restore approximal caries lesions must be based on the accurate assessment of

Saulo Geraldeli, DDS, MS, PhD, associate professor, Department of General Dentistry, Division of Biomedical Materials, East Carolina University, School of Dental Medicine, Greenville, NC, USA

Patricia NR Pereira, DDS, PhD, clinical associate professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Marc E Ottenga, DDS, clinical professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Deborah A Dilbone, DMD, associate professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

\*Corresponding author: 1395 Center Dr, Room D9-6, PO Box 100415, Gainesville, FL 32610-0415; e-mail: mnascimento@dental.ufl.edu

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<sup>\*</sup>Marcelle M Nascimento, DDS, MS, PhD, associate professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Ana P Ribeiro, DDS, MS, PhD, clinical assistant professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Alex J Delgado, DDS, MS, clinical associate professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Luisa Cassiano, DDS, MS, clinical assistant professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Maria GD Caraballo, DDS, adjunct clinical assistant professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

Jean-François Roulet, Dr. med. Dent., Dr. h.c., professor, Department of Restorative Dental Sciences, Division of Operative Dentistry, College of Dentistry, University of Florida, Gainesville, FL, USA

tooth cavitation, as the accumulation of oral biofilms in these areas encourages lesion progression. However, lesions radiographically into dentin remain the main threshold criterion for restoring approximal lesions even though most of these lesions may not be cavitated. A school-based clinical protocol for temporary tooth separation (TTS) was developed to improve visual-tactile assessment and management of clinically inaccessible approximal lesions. TTS data retrieved from electronic health records were used to correlate radiographic lesion depth and surface cavitation status with lesion location and the patient's caries risk and to evaluate the effectiveness of TTS as a diagnostic aid for approximal lesions. Of the 206 lesions assessed, 66.5% (n=137) were located in the maxillary arch, 56.6% (n=116) in distal surfaces, 61.3% (n=114) in premolars, and 21.5% (n=40) in molars. After tooth separation, 79.6% (n=164) of the lesions were diagnosed as noncavitated, including 90% (n=66) of the lesions radiographically at the inner half of enamel (E2) and 66% (n=49) of those at the outer-third of dentin (D1). Logistic regression analysis using E2 and D1 lesions showed no significant association between lesion depth or cavitation status with lesion location and caries risk. TTS is a feasible and effective diagnostic aid for the assessment and appropriate management of approximal caries lesions. There is a need to reevaluate the use of radiographic lesion depth as the single determinant of the restorative threshold for clinically inaccessible approximal lesions.

#### INTRODUCTION

Substantial knowledge of the caries process based on over 100 years of caries research supports the use of preventive, risk-based, and tooth-preserving approaches for caries management.1 Caries is well known as a biofilm-mediated disease. That is, tooth cavitation or any situation in the oral environment that encourages the accumulation and metabolic activity of oral biofilms (dental plague) increases the risk for caries development and progression. Efforts to arrest caries activity using biofilm control and remineralization therapy must take place at all times but especially prior to enamel cavitation.<sup>3-5</sup> Remineralization therapy aims to stop or reverse the progression of caries lesions by combining the use of fluoride products, education, and behavioral changes that promote mineralization over demineralization of dental tissues.<sup>2</sup> In contrast to the outdated and traditional surgical-only approach, the current consensus is that enamel and noncavitated carious lesions that can be arrested and remineralized by nonsurgical approaches should not be restored, whereas noncleansable and cavitated lesions progressing into dentin most often require restorative intervention.<sup>6,7</sup> In particular, the appropriate time to restore approximal lesions is when the lesions are cavitated because they are not cleansable or amenable to the effects of fluoride, and therefore these lesions are more likely to progress.

The decision to restore approximal caries lesions must be based on the accurate assessment of the presence of tooth cavitation. However, visual-tactile assessment of cavitation of these surfaces can be challenging due to the restricted access to contacting surfaces. Consequently, bitewing radiography has remained the primary detection method for clinically inaccessible approximal lesions. Since the probability of cavitation increases with lesion depth, lesion progression into the outer half of dentin, or D1 lesions, has become the threshold criterion for surgical (restorative) intervention of approximal lesions. 1,2 While bitewing radiographs are useful for detecting tooth demineralization, they may not reveal the surface integrity with accuracy.8-10 In fact, earlier clinical studies revealed that between 40% and 60% of D1 lesions are not cavitated. 8,9,11-13 These findings imply that restorations leading to irreversible loss of tooth structure are commonly placed in teeth with noncavitated approximal lesions based on an age-old assumption that all caries lesions will progress if left untreated and not on accurate assessment of surface cavitation. Of greater concern, dentists' restorative thresholds remain unaffected by scientific evidence supporting less invasive management of noncavitated caries lesions. A systematic review shows that 21% of dentists or dental therapists would intervene restoratively on approximal lesions radiographically confined to enamel and that 48% would intervene restoratively on approximal lesions extending up to the dentinenamel junction.<sup>6</sup>

The use of elastic orthodontic separators has long been proposed to achieve interproximal tooth separation and improve visual-tactile assessment of approximal tooth surfaces. 1,8-10,14 Unfortunately, this technique remains as an elective procedure that is not commonly used in clinical practice. Tooth separation allows the clinical evaluation of surface integrity (absence or presence of a microcavity or a distinct cavity) and lesion activity. A school-based

clinical protocol on temporary tooth separation (TTS) was developed to aid in direct visual-tactile assessment, thus facilitating accurate diagnosis and appropriate management of approximal caries lesions with an emphasis on preventive and minimally invasive restorative treatment planning based on the status of individual tooth surfaces. This study used electronic health records (EHR) of the TTS data to examine: a) the correlations between radiographic lesion depth, surface cavitation status, lesion location, and patients' caries risk, and b) the potential value of TTS as a diagnostic aid for approximal caries lesions.

#### **METHODS AND MATERIALS**

#### **Study Design**

This retrospective study used axiUm (Software Advice, Inc) electronic reports generated from patients' EHR to collect data on different variables related to the procedure of TTS, including radiographic lesion depth (outer [E1] or inner [E2] half of enamel, and outer [D1], middle [D2], or inner [D3] third of dentin), surface cavitation status (cavitated or noncavitated), lesion location (mesial/distal, maxillary/mandibular arch, and tooth type), and patients' caries risk level (extreme, high, moderate, or low). Data were collected for the period from August 29, 2017, to July 30, 2019.

#### **Clinical Procedures**

The procedures of clinical examination, radiographic interpretation, caries risk assessment, TTS, and development of treatment plans were performed by third- and fourth-year predoctoral dental students under the supervision of the attending clinical faculty from the Department of Restorative Dental Sciences. Students received didactic and preclinical training on diagnosis, prevention, and management of dental caries during their first two years of the DMD program. Faculty meetings and calibration sessions were conducted to discuss the scientific evidence available regarding caries diagnosis and management, which included the development and implementation of the TTS protocol. The school's caries risk assessment and management program was developed based on the Caries Management by Risk Assessment (CAMBRA) system as described elsewhere. 15

#### **Protocol for TTS**

The indications and steps of the clinical and electronic-based protocol for TTS are shown in Figure 1. According to the protocol, TTS must be

performed for the assessment of all approximal caries lesions showing no evident cavitation on visual-tactile examination and extending to the outer third of dentin (D1 lesions) upon radiographic examination. However, TTS is also recommended for other types of radiographic lesion depth when the lesion activity and/or surface cavitation are questionable. For the lesions assessed during the study period, the orthodontic bands were placed in the interproximal spaces, and TTS was performed for at least one hour but no longer than three hours. The protocol recommends the placement of bands as early in the dental appointment as possible to allow more time for tooth separation and thus achieve adequate visual-tactile assessment of the tooth surfaces. Of note, multiple lesions could have been assessed on the same patient during one or several dental appointments.

The TTS electronic-based protocol was developed and built into axiUm. The protocol consists of using an axiUm procedure code (D0426) and a form (DEPCAV) that are entered in each patient's EHR for each tooth surface assessed with tooth separation. The D0426 code was used to record the separation procedure and the tooth number and surface. The DEPCAV form was populated, linked to D0426, and used to provide further diagnostic information on radiographic depth and cavitation status (as determined clinically after tooth separation) of the caries lesions. Another protocol was developed to monitor active approximal caries lesions that had been assessed as noncavitated after tooth separation. The D0604 code was used to record the monitoring procedure and tooth number and surface, while the CMPLAN form (linked to D0604) was used to provide information on the patient's caries risk, radiographic lesion depth, time frame for recall visits, and the procedures to be performed at recalls (Supplemental Material 1).

#### **Diagnosis and Management of Caries Lesions**

After tooth separation, the approximal caries lesions were directly assessed and diagnosed by visual-tactile examination using the criteria shown on Table 1, which also shows the treatment recommendations according to the surface cavitation status (surface integrity) and activity of the caries lesions. Lesion activity was determined by clinical appearance and texture of tooth surfaces. Even though Table 1 shows that the color of tooth surfaces varied according to the type of dental tissue affected and lesion activity, color was not to be used as a



Figure 1. Tooth separation and assessment of the mesial caries lesion of tooth 29; (a) and (b): Initial views of the orthodontic elastic separator between teeth 28 and 29 that remained in place for 90 minutes. (c) and (d): Clinical aspect after separation.

Clinical and electronic-based protocol for temporary tooth separation. Indication: Temporary tooth separation must be performed for the assessment of all approximal caries lesions showing no evident cavitation on visual-tactile examination and extending to the outer third of dentin (D1 lesions) on radiographic examination. Clinical Steps:

- Inform the patient about the need, indications, advantages, and disadvantages of the tooth separation procedure.
- Stretch the orthodontic bands outside of the mouth by using an orthodontic forceps or dental floss ligatures. There are two different sizes of orthodontic bands available in our clinics. Whenever possible, the tighter bands and perhaps two bands should be inserted into the interproximal spaces in order to achieve satisfactory tooth separation.
- Work the orthodontic bands down through the contact point. The bands should be left interproximally for at least one hour, but longer separation time typically yields better results.
- After the separation time, remove the bands gently by using cotton plyers, dental explorer, or dental floss.
- 5. Wash and dry the approximal tooth surfaces.
- 6. When the teeth are well separated, you should be able to visualize the tooth surfaces directly or indirectly (mirror) and, if needed, to "feel" the surface's texture by moving the tip of a dental explorer gently across the tooth surfaces.
- 7. Determine if caries lesions are present on the approximal surfaces. If the surfaces present no signs of past or present caries activity, this must be noted on the patient's progress notes, and no further treatment is necessary. If caries lesions are present, you must assess surface integrity (cavitated or noncavitated) and lesion activity (active or inactive).
- For active and noncavitated carious lesions, fluoride varnish must be applied while the teeth are still separated. These lesions will be managed by nonsurgical procedures and monitored over time.
- All clinical observations, caries diagnoses, and corresponding treatment plans must be identified and recorded in the patient's dental record.

diagnostic characteristic to distinguish active from inactive lesions.

According to the protocol, and regardless of the patient caries risk, enamel and D1 lesions assessed as noncavitated active lesions were to be managed by nonsurgical procedures and monitored over time, while restorative treatment was to be planned for cavitated active lesions. At this point, the treatment of the noncavitated lesion was to be based on controlling patient's risk factors through therapeutic means and lifestyle changes. To stop the progression of the disease process, the focus was to be on the use of fluoride (medication), the quality and frequency of toothbrushing/flossing (plaque control), and dietary changes (lifestyle). The lesion was to be identified in the patient's EHR, the proper monitoring codes entered into axiUm, and an appropriate recall time for reassessment created.

#### **Statistical Analysis**

Data management and statistical analyses were performed using Stata SE 15.0 at a significance level of 0.05. For descriptive analysis, the distribution of percentages and means were calculated when appropriate. A normality test was used to determine if the data set was well modeled by a normal distribution. Given the fact that most dentists use the restorative threshold of radiographic D1 or even E2 lesions, a logistic regression model was used to correlate radiographic depth, cavitation status, lesion location, and patients' caries risk level for a total of 134 E2 and D1 lesions from 64 patients whose caries risk had been assigned in the EHR. The logistic regression model used caries risk (patient level), type of arch (maxillary/mandibular), tooth surface (mesial/distal), and tooth (incisor, canine, premolar or molar) as independent variables at the levels of lesion and patient. Hence, the model accounted for teeth from the same patient entering the patient in the logistic regression as a variable.

#### **RESULTS**

A total of 206 approximal caries lesions from 97 patients (mean average of 2.1 lesions; range of 1 to 16 lesions per patient) were assessed following the protocol for TTS, and information on these lesions was electronically recorded and retrieved for the study analysis. The distribution of these lesions according to location were as follows: 116 (56.6%) were located on distal surfaces and 90 (43.9%) on mesial surfaces; 114 (61.3%) were located on premolars, 40 (21.5%) on molars, 28 (15.1%) on incisors, and 24 (11.7%) on canines; and 137 (66.5%) were

	Diagnostic Characteristics	Treatment						
Dental Tissue	Appearance/Texture	Surface Integrity	Activity	Recommendations				
Enamel	Whitish/yellowish opacity     Shiny surface     Smooth surface upon gentle probing     May exhibit localized shallow defects or microcavitations	Noncavitated	Inactive	<ul> <li>Review oral hygiene instructions, diet, and the use of fluoride</li> <li>Monitor lesion progression over time</li> </ul>				
Enamel	Whitish/yellowish opacity     Loss of luster; 'chalky' or 'milky' appearance     Rough surface upon gentle probing     May exhibit localized shallow defects or microcavitations	Noncavitated	Active	<ul> <li>At the tooth separation visit, apply fluoride varnish while teeth are still separated</li> <li>Review oral hygiene instructions, diet, and the use of fluoride</li> <li>Use of at-home fluoride products as part of regular oral hygiene</li> <li>In-office fluoride varnish applications until lesion activity is controlled (arrested)</li> <li>Monitor lesion progression over time</li> </ul>				
Dentin	Brown or black color     Shiny surface     Hard surface upon gentle probing	Cavitated	Inactive	<ul> <li>Review oral hygiene instructions, diet, and the use of fluoride</li> <li>Restorative treatment only if compromising the tooth function and the control of lesion activity</li> </ul>				
Dentin	Yellow to brown color     Surface breakdown     Soft or leathery texture of exposed dentin	Cavitated	Active	Review oral hygiene instructions, diet, and the use of fluoride     Restorative treatment				

<sup>a</sup> The color of an inactive lesion may vary from white to brown or black, but color should not be used as a diagnostic characteristic to distinguish active from inactive

noncavitated lesions. All treatment recommendations must be accompanied by the management of the patient's caries risk factors.

located in the maxillary arch and 69 (33.5%) in the mandibular arch.

Table 2 shows the distribution of the lesions according to their radiographic depth and cavitation status. Of the lesions assessed by TTS, 47 (22.8%) were E1, 73 (35.4%) were E2, 74 (35.9%) were D1, and 12 (5.8%) were D2 lesions as determined by radiographic examination prior to TTS. No D3 lesion was assessed by TTS during the study period. After TTS, 92% (n=43) of E1, 90% (n=66) of E2, 66% (n=49) of D1, and 50% (n=6) of D2 lesions were assessed as noncavitated. The logistic regression model showed no significant association between lesion radiographic depth (p>0.05) or presence of cavitation (p>0.05) with

any of the independent variables tested, as shown in Table 3.

#### DISCUSSION

The most significant findings of this study were that the majority of approximal lesions extending radiographically up to the dentin-enamel junction (E2) and those at the outer third of dentin (D1) were assessed and diagnosed as noncavitated after TTS. Importantly, E2 and D1 lesions are the most likely type of lesions to be restored by dentists when using radiographic examination as the primary method to determine restorative needs. Current evidence supports that treatment strategies should be aimed at arresting carious lesions with intact tooth surfac-

Table 2:	Distribution of Caries Lesions by Radiographic Depth and Surface Cavitation Status as Determined Before and After
i	Tooth Separation, Respectively <sup>a</sup>

•						
	E1	E2	D1	D2	D3	Total
	5		5	5	9	
Noncavitated, no. (%)	43 (91.5)	66 (90.4)	49 (66.2)	6 (50)	0	164 (79.6)
Cavitated, no. (%)	4 (8.5)	7 (9.6)	25 (33.8)	6 (50)	0	42 (20.4)
Total, no. (%)	47 (100)	73 (100)	74 (100)	12 (100)	0	206 (100)
2						

<sup>&</sup>lt;sup>a</sup> The radiographic depth of caries lesions were recorded as being at the outer (E1) or inner (E2) half of enamel or at the outer (D1), middle (D2), or inner (D3) third of dentin. Percentages are with columns for each lesion depth.

Table 3: Logistic Regression Models for Radiographic Lesion Depth and Presence of Surface Cavitation<sup>a</sup>

Variables	Coefficient	<i>p</i> -Value	95% Confidence Interval								
Radiographic depth (enamel/dentin)											
Caries risk	0.0767299	0.812	-0.55-0.71								
Patient	-0.0125629	0.235	-0.03-0.01								
Arch	0.452156	0.574	-1.12-2.03								
Tooth type	0.1689549	0.329	-0.17-0.51								
Tooth surface	-0.0299411	0.509	-0.12-0.06								
Presence of cavita	ation (yes/no)										
Caries risk	-0.1225252	0.755	-0.89-0.65								
Patient	0.0035214	0.785	-0.022-0.03								
Arch	0.442721	0.656	-1.50-2.39								
Tooth type	-0.2908966	0.143	-0.68-0.10								
Tooth surface	0.0352046	0.526	-0.07-0.14								

<sup>&</sup>lt;sup>a</sup> The logistic regression model used caries risk (extreme, high, moderate, or low), type of arch (maxillary or mandibular), tooth type (incisor, canine, premolar, or molar), and tooth surface (mesial or distal) as independent variables at the levels of lesion and patient.

es and that restorative intervention should be considered only for cavitated lesions. Based on existing literature, it is not surprising that most E1 lesions were diagnosed as noncavitated. However, the high prevalence of E2 (90%), D1 (66%), and even D2 (50%) lesions diagnosed as noncavitated supports the value of placing orthodontic separators as an effective diagnostic aid for approximal caries lesions. Here, irreversible loss of tooth structure by restorative treatment was prevented in a considerable number of caries lesions based on the accurate assessment of their cavitation status.

Advantages of the TTS technique include low-cost, fast, temporary with reversible separation and the ability to diagnose approximal lesions with sufficient accuracy to support appropriate treatment decisions. Disadvantages may include the fact that occasionally the teeth are not as accessible as needed after the separators are placed, and that the use of the separators may create some brief and mild discomfort for patients. Another disadvantage of this technique is the requirement for an extra office visit if separators are used for longer than 24 hours. While the use of separators for longer periods of time may result in better tooth separation of approximal tooth surfaces, reasonable separation (sufficient to improve assessment) can be achieved when separators are used for a minimum of one hour as described on this study. As this study was performed in an educational setting, further considerations regarding time and work flow must take place when implementing TTS protocols in private and public practice settings. TTS can be performed at any dental visit when the assessment of approximal caries lesions is required, and the separators can remain in the interproximal space while other dental procedures are being performed so as to not disrupt the sequence of the treatment plan. Ideally, the placement of orthodontic bands in the interproximal spaces should be performed as early in the dental appointment as possible to allow more time for tooth separation. If preferable, tooth separators can also remain in place for 24 to 48 hours, but a second dental visit will be required for assessment and removal of the orthodontic bands.

The development and validation of clinical practice protocols or guidelines are necessary to disseminate the best evidence currently available, reduce variability in practice patterns, improve the quality of patient care, and reach consensus among practitioners. <sup>16</sup> A survey-based study revealed that US dental schools use a variety of different approaches to evaluate, select, and implement evidence-based clinical guidelines endorsed by the American Dental Association, and that very few schools were shown to have an effective set of policies and procedures to support guideline implementation. <sup>16</sup>

Undoubtedly, the academic routes to introduce new clinical protocols in an educational setting can have several impediments prior to actual and effective implementation. The development of the TTS protocol described here included the testing of technique feasibility and patients' acceptance in our predoctoral dental clinics, and both were proven acceptable. Notably, patients tolerated the procedure well and were pleased when they were informed that their teeth did not have a cavity and that no "drilling and filling" were needed.

The use of EHR can facilitate careful documentation of the diagnosis, treatment plan, and monitoring plans for caries lesions. Our electronic-based protocol for TTS was developed using axiUm, but it is anticipated that a similar protocol can be developed and adapted for other software programs. The use of our electronic-based protocols and tracking systems can certainly facilitate data collection to support evidence on the use of TTS as a diagnostic aid for approximal caries lesions, and they may also contribute to a reexamination of the optimal threshold for restorative intervention on approximal lesions. This information will also be useful to track compliance and to provide feedback to students and faculty about their own personal implementation of the clinical protocol.

Caries risk assessment is key to the success of the caries management plan. However, according to our TTS protocol, treatment recommendations for approximal caries lesions are based on lesion cavitation status and activity as determined by the TTS procedure and shown in Table 1, and not based on the patient's caries risk level. Treatment recommendations should also take into consideration the patients' needs, risks, motivation and compliance capabilities. Of great importance, dental care providers should explain the treatment options for individual lesions (nonsurgical and/or surgical approaches) to patients and provide the necessary educational resources for patients to choose the treatment option that best aligns with their unique cultural and personal beliefs and lifestyles. Effective patient participation and engagement in a shared decision-making process certainly support the compliance and promotion of dental health. Regardless of the treatment decision, there must be a rigorous and specific strategy in place for assessing and monitoring caries activity and lesion progression over time. It is even more critical to closely monitor the activity of noncavitated approximal lesions to evaluate whether the management recommendations based on nonsurgical approaches have been effective at arresting these lesions. Diagnostic codes and electronic forms were developed to create a monitoring plan for active approximal caries lesions that were assessed as noncavitated after tooth separation. At each recall visit, lesion activity and cavitation status must be reassessed, at-home preventive recommendations should be reinforced, and treatment approaches should be reevaluated as needed. Effective patient participation and engagement in a shared decision-making process will certainly facilitate compliance with treatment recommendations and successful promotion of dental health.

Although robust evidence exists supporting risk-based and tooth-preserving approaches for caries management, evidence for long-term clinical outcomes of noncavitated lesions assessed by tooth separation is lacking. Only one longitudinal study reported a range of 20% to 44% in cavitation prevalence of dentin lesions after two years of recall visits (six-month interval) but with limited preventive intervention. It can be argued that the scientific basis for many of the "newer" caries management approaches is not yet validated, but neither is the scientific basis for many of the surgical procedures done in dentistry. As previously discussed in different venues, when the well-being of

the patient is considered, it is more important to carry out a risk-based caries management plan incorporating the best available evidence than to do nothing due to the lack of strong evidence.<sup>6, 18</sup>

#### CONCLUSIONS

Findings from this study support that the use of temporary tooth separation is in line with the tooth-preserving philosophy of modern caries management, and that it is a feasible and effective diagnostic aid for assessment and appropriate management of approximal caries lesions. There is a clear need to reevaluate the use of radiographic lesion depth as the single determinant of the restorative threshold for clinically inaccessible approximal lesions.

#### Acknowledgements

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#### **Regulatory Statement**

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of the Institutional Review Board of the University of Florida. The approval code issued for this study is IRB201800458.

#### **Conflict of Interest**

The authors of this article certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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# Laser Influence on Dental Sensitivity Compared to Other Light Sources Used During In-office Dental Bleaching: Systematic Review and Meta-analysis

BGS Casado • EP Pellizzer • JR Souto Maior • CAA Lemos • BCE Vasconcelos • SLD Moraes

#### Clinical Relevance

The use of laser light during bleaching will not reduce the incidence or severity of sensitivity and will not increase the degree of color change compared with nonlaser light sources

#### **SUMMARY**

Objective: To evaluate whether the use of laser during in-office bleaching promotes a reduction in dental sensitivity after bleaching compared with other light sources.

Methods: The present review was conducted following the Preferred Reporting Items for Systematic Reviews and Meta-analyses (PRISMA) and is registered with PROSPERO (CDR42018096591). Searches were conducted in the PubMed/Medline, Web of Science, and

Cochrane Library databases for relevant articles published up to August 2018. Only randomized clinical trials among adults that compared the use of laser during in-office whitening and other light sources were considered eligible.

Results: After analysis of the texts retrieved during the database search, six articles met the eligibility criteria and were selected for the present review. For the outcome dental sensitivity, no significant difference was found favoring any type of light either for intensity

Belmiro Cavalcanti do Egito Vasconcelos, PhD, DDS, associate professor, Dentistry School, University of Pernambuco, Recife, Brazil

Sandra Lucia Dantas de Moraes, PhD, DDS, adjunct professor, Dentistry School, University of Pernambuco, Recife, Brazil

\*Corresponding author: Av General Newton Cavalcanti, 1650, Tabatinga, Camaragibe-PE, Brazil, 54.756-220; e-mail: brunocasado@hotmail.com

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<sup>\*</sup>Bruno Gustavo da Silva Casado, PhD, DDS, adjunct professor, Dentistry School, University of Pernambuco, Recife, Brazil

Eduardo Piza Pellizzer, PhD, DDS, full professor, Araçatuba Dental School, São Paulo State University (UNESP). Araçatuba, Brazil

Juliana Raposo Souto Maior, PhD, DDS, Dentistry School, University of Pernambuco, Recife, Brazil

Cleidiel Aparecido Araújo Lemos, PhD, DDS, Araçatuba Dental School, São Paulo State University (UNESP). Araçatuba. Brazil

(mean difference [MD]: -1.60; confidence interval [CI]: -3.42 to 0.22; p=0.09) or incidence (MD: 1.00; CI: 0.755 to 1.33; p=1.00). Regarding change in tooth color, no significant differences were found between the use of the laser and other light sources (MD: -2.22; CI: -6.36 to 1.93; p=0.29).

Conclusions: Within the limitations of the present study, laser exerts no influence on tooth sensitivity compared with other light sources when used during in-office bleaching. The included studies demonstrated that laser use during in-office bleaching may have no influence on tooth color change.

#### INTRODUCTION

Tooth whitening is one of the most common cosmetic procedures performed in dental offices due to the fact that this conservative, easy-to-execute method offers fast results. Among the different forms of treatment available for bleaching vital teeth, the in-office method is an alternative. This clinical method involves the direct supervision by a dental professional throughout the application of the bleaching agent (hydrogen peroxide) to the dental surfaces. 2,3,5,6

Hydrogen peroxide serves as a strong oxidizing agent, leading to the formation of free radicals.<sup>5</sup> These free radicals attack complex pigment molecules, resulting in smaller, colorless by-products that establish the whitening effect seen in the enamel.<sup>1,7-9</sup>

In the attempt to enhance the bleaching effect, many manufacturers recommend the use of light or heat for the catalytic decomposition of the peroxides. <sup>1,6,10</sup> Several sources of light proposed for this purpose include lasers, light-emitting diodes (LEDs), plasma arc lamps, and halogen lamps. <sup>6,11,12</sup> However, this practice is risky as studies show that the use of light sources can have side effects, such as tooth sensitivity, due to the increase in intrapulpal temperature and the influx of free radicals from the hydrogen peroxide, which can penetrate the dental structure and reach the pulp. <sup>13-15</sup>

On the other hand, some authors report that the use of laser in combination with in-office bleaching agents is recommended as an adequate strategy to reduce dental sensitivity after office bleaching as this source emits well-defined monochromatic light at a single wavelength, thereby reducing the risk of increasing intrapulpal temperature. Laser has also been claimed to have analgesic properties. <sup>19</sup>

Thus, the aim of the present systematic review and meta-analysis was to evaluate whether the use of a laser during in-office dental bleaching promotes a reduction in tooth sensitivity after bleaching compared with other light sources. The hypothesis was that the application of laser during in-office dental bleaching reduces tooth sensitivity after bleaching.

#### **METHODS**

#### **Registration Protocol**

The present systematic review was conducted in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-analyses (PRISMA statement).<sup>20</sup> The methods used for this systematic review have been registered with PROSPERO (CDR42018096591).

#### Eligibility Criteria

A specific question was developed based on the PICO (population, intervention, control, and outcomes) criterion. The following was the guiding question: Does the use of laser during in-office dental bleaching promote a reduction in tooth sensitivity and color change compared with other light sources? The population was composed of patients receiving in-office bleaching using a light source; the intervention was in-office bleaching combined with the use of the laser, and the control was in-office bleaching with other light sources (LED, halogen light, and plasma arc lamps). The primary outcome was tooth sensitivity after bleaching, and the secondary outcome was the change in color.

Prospective randomized clinical trials published in the English language that had at least 10 patients and made a direct comparison between the use of laser during in-office whitening and other light sources were considered eligible. Retrospective studies, crossover studies, *in vitro* studies, *in situ* studies, animal studies, mechanical studies, case reports and reviews of the literature were excluded.

#### Search Methods and Search Strategy

Two independent reviewers (BGSC and CAAL) performed searches in the Cochrane Library, Pubmed/MEDLINE, and Web of Science databases for relevant articles published up to August 2018. The following search terms were used: "dental bleaching and light OR tooth bleaching and light OR teeth bleaching and light OR dental whitening and light OR tooth whitening and light OR teeth whitening and light."

The first phase of the selection process involved an analysis of the titles and abstracts retrieved during the search of the electronic databases. For studies in was not possible to obtain sufficient information, the complete article was obtained. The two researchers also performed a manual search for articles published up to August 2018 in specific journals in the field: Dental Materials, Journal of Dentistry, Operative Dentistry, and Clinical Oral Investigations. A manual search of the OpenGrey database (www.opengrey.eu) was also conducted to determine relevant articles in the grey literature. A third reviewer (JRSM) examined all the divergences of opinion between the two reviewers regarding the selection of articles, and a consensus was reached through discussion.

#### Risk of Bias and Evaluation of Study Quality

Two reviewers (BGSC and CAAL) evaluated the methodologic quality of the studies included in the review. For such, the Cochrane criteria were used to appraise the quality and risk of bias based on sequence generation, allocation concealment, blinding of participants, personnel or outcome investigator, incomplete outcome data, selective outcome reporting, and other sources of bias. Using the Cochrane system, the risk of bias is classified as low, high, or unclear.

#### **Data Extraction**

Data were extracted by one of the reviewers (BGSC) to tabulate data of greater relevance to the analysis of the studies. The following data were extracted: author/year, type of study, patients, type of light used, bleaching agent/concentration, tooth sensitivity, and tooth discoloration.

#### **Meta-analysis**

The meta-analysis was based on the Mantel-Haenzel and the inverse variance methods using Review Manager 5 (Cochrane Group, Haymarket, London, United Kingdom). The dichotomous outcome (absolute risk of tooth sensitivity) was evaluated using the risk ratio, and continuous outcomes (intensity of tooth sensitivity, subjective and objective color change) were evaluated based on the mean difference. A p-value <0.05 was considered indicative of statistical significance. Heterogeneity was evaluated based on the  $I^2$  value (25%=low, 50%=moderate, and 75%=high). When heterogeneity was statistically significant (p<0.10), a random-effects model was used for the meta-analysis; otherwise, a fixed-effects model was used.

#### **Additional Analysis**

An additional analysis was performed using the Kappa coefficient to establish interexaminer agreement in the selection process of the studies from the three databases. The Kappa value was obtained by evaluating the titles and abstracts selected. High levels of agreement were found for the Cochrane Library (K=0.86), PubMed/MEDLINE (K=0.88), and Web of Science (K=0.80) databases.

#### **RESULTS**

The search of the databases led to the retrieval of 1301 articles: 763 from PubMed/MEDLINE, 395 from Web of Science, and 143 from the Cochrane Library. After duplicates were removed, 995 titles and abstracts were analyzed based on the eligibility criteria. Ten studies were preselected for full-text analysis, of which four were excluded for the following reasons: literature review, failure to use the laser as a light source, and failure to compare laser to other light sources. Details on the search strategy are presented in the flowchart (Figure 1).

Six studies were selected for data analysis and are listed in detail in Table 1. All studies were randomized controlled trials, one of which had a split-mouth design. A total of 136 patients were included in the studies. Sample sizes ranged from 16 to 40 individuals age 18 to 70 years. In all studies, hydrogen peroxide was the in-office bleaching agent used, with a concentration of 35% in three studies, 37% in one study, and 38% in two studies. Evaluations of dental sensitivity were performed in all studies. Four studies evaluated the incidence of sensitivity and three evaluated intensity using the visual analog scale. In four studies, the change in tooth color was evaluated subjectively using a color scale.

#### **Risk of Bias**

Each trial was assessed for risk of bias; the scores are displayed in Figure 2. A low risk of bias was found regarding participant blinding, staff assessment and results, incomplete outcome, and selective reporting items. However, all studies<sup>22-27</sup> had unclear bias regarding the random sequence generation and allocation concealment.

#### **Meta-analysis**

Tooth Sensitivity—Dental sensitivity was evaluated in two ways: intensity and incidence. Intensity was determined when the patient quantified his or her response to pain on a visual analog scale, while



#### **PRISMA 2009 Flow Diagram**

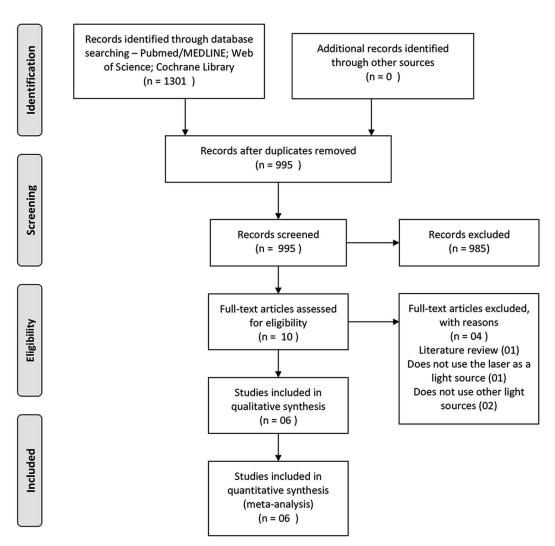


Figure 1. Flowchart showing steps of article selection process.

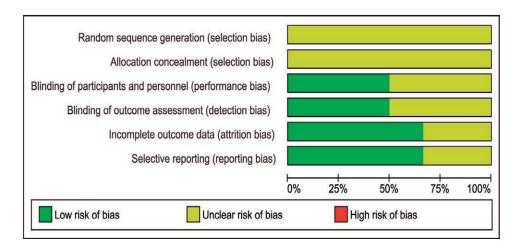


Figure 2. Cochrane scale for bias

Author, Stud		Sample	Sex	Light	Bleaching	Light	Tooth Se	ensitivity	Color	Change
Date		Size	Size Source (N) Ager		Agent	Source Time per Session	Intensity	Incidence	Subjective Shade Evaluation	Objective Shade Evaluation
Marson and	RCT	30	NR	Halogen light (10)	35% HP	NR	NR	5 (10)	NR	NR
others, 2008 <sup>22</sup>				LED (10)			NR	8 (10)		
				LED/Laser (10)			NR	6 (10)		
Gurgan and others, 2010 <sup>23</sup>	RCT	30	M: 11 (27.5%) F: 29 (72.5%)	LED (10)	38% HP	1st 20′ 2nd 20′	2.9±1.48	NR	8.5±3.59	5.43±0.20
				Laser (10)	37% HP	$1st 7 \times 15''$ $2nd 4 \times 15''$ $3rd 4 \times 15''$	0.59±0.92	NR	8.6±3.19	5.59±0.172
				PAC (10)	•	1st 7' 2nd 7' 3rd 7'	3.8±1.29	NR	8.4±2.98	5.28±0.096
de Almeida and others, 2012 <sup>24,25</sup>	RCT	20	NR	Halogen light (10)	35% HP	1st 20" 2nd 20" 3rd 20"	2.98±2.97	10 (10)	1.1±0.31	NR
				LED/laser (10)	•	1st 3' 2nd 3' 3rd 3'	2.2±3.22	10 (10)	1.0±0.1	NR
Polydorou, and others 2013 <sup>26</sup>	RCT	40	NR	Halogen light (10)	38% HP	1st 30" 2nd 30" 3rd 30" 4th 30"	NR	NR	5.5±2.94	6.1±1.9
				Laser (10)	•	1st 8' 2nd 8' 3rd 8' 4th 8'	NR	NR	1.15±1.7	2.15±2.4
Farhat and others,	RCT (SM)	16	NR	LED (16)	35% HP	1st 3'	0.61±1.17	6 (16)	2.75±1.53	NR
2014 <sup>27</sup>				LED/laser (16)	•	2nd 3' 3rd 3'	0.75±1.2	7 (16)	2.63±1.02	NR

Abbreviations: F, female; HP, hydrogen peroxide; LED, light-emitting diode; M, male; NR, not reported; PAC, plasma arc lamp; RCT, randomized controlled trial; SM, split mouth.

incidence was assessed by the number of sensitivity events.

The intensity of dental sensitivity was measured using the visual analog scale (continuous outcome) in three studies. The random-effects model revealed no significant difference favoring any type of light (mean difference [MD]: -1.60; confidence interval [CI]: -3.42 to -0.22; p=0.09). Moreover, laser did not lead to significantly lower dental sensitivity compared with the other light systems (Figure 3). However, the data were heterogeneous ( $\chi^2$ : 29.39;  $I^2=90\%$ ; p<0.0001) and the studies included in the analysis did not share a common effect size.

The incidence of dental sensitivity among the light systems combined with the in-office bleaching based on visual analog scale scores was evaluated in three studies. The relative risks revealed no differences between laser and the other light sources (MD: 1.00; CI: 0.755 to 1.33, p=1.00). Heterogeneity was nonsignificant (Figure 4), and all studies included in the analysis shared a common effect size,

suggesting no differences in the incidence of dental sensitivity among the light sources used.

Color Change—The effectiveness of whitening was determined in three studies using the subjective method (Vita Classical shade guide) immediately after dental whitening. The random-effects model revealed no significant differences between the use of laser and other light sources combined with inoffice dental bleaching (MD: -2.22; CI: -6.36 to 1.93; p=0.29). The data were heterogeneous ( $\chi^2$ : 30.60;  $I^2=97\%$ ; p<0.00001) (Figure 5), and the studies included in the analysis did not share a common effect size.

#### DISCUSSION

In the present systematic review and meta-analysis, no significant differences were found in the reduction of dental sensitivity (intensity or incidence) or the change in tooth color when the use of the laser was compared with other light sources (LED, halogen light, and plasma arc lamps) during in-

#### Forest plot Intensity

	Expe	rimen	tal	С	ontro	1		Mean Difference	Mean Difference
Study or Subgroup	Mean	SD	Total	Mean	SD	Total	Weight	IV, Random, 95% CI	IV, Random, 95% CI
Almeida et al 2012 (Halogen llight)	2.2	3.22	10	2.98	2.97	10	17.9%	-0.78 [-3.50, 1.94]	-
Farhat et al., 2014 (LED)	0.75	1.2	16	0.61	1.17	16	28.0%	0.14 [-0.68, 0.96]	*
Gurgan et al 2010 (LED)	0.59	0.92	10	2.9	1.48	10	26.9%	-2.31 [-3.39, -1.23]	*
Gurgan et al., 2010 (Plasma Arc Lamp)	0.59	0.92	10	3.8	1.29	10	27.3%	-3.21 [-4.19, -2.23]	*
Total (95% CI)			46			46	100.0%	-1.60 [-3.42, 0.22]	•
Heterogeneity: Tau <sup>2</sup> = 2.90; Chi <sup>2</sup> = 29.39	df = 3 (	< 0.0	0001);	I2 = 909	%			-	-20 -10 0 10 20
Test for overall effect: Z = 1.72 (P = 0.09)	)								Favours [experimental] Favours [control]
3									ravous (experimental) Pavous (control)

#### Forest plot Incidence

	Experim	ental	Contr	rol		Risk Ratio	Risk Ratio
Study or Subgroup	<b>Events</b>	Total	<b>Events</b>	Total	Weight	M-H, Fixed, 95% CI	M-H, Fixed, 95% CI
Almeida et al 2012 (Halogen llight)	10	10	10	10	35.6%	1.00 [0.83, 1.20]	- <del>+</del> -
Farhat et al., 2014 (LED)	7	16	6	16	20.3%	1.17 [0.50, 2.71]	<del>-  •</del>
Marson et al 2008 (Halogen light)	6	10	5	10	16.9%	1.20 [0.54, 2.67]	<del></del>
Marson et al 2008 (LED)	6	10	8	10	27.1%	0.75 [0.41, 1.36]	
Total (95% CI)		46		46	100.0%	1.00 [0.75, 1.33]	<b>*</b>
Total events	29		29				
Heterogeneity: Chi <sup>2</sup> = 1.23, df = 3 (P	= 0.75); 12	= 0%					0.2 0.5 1 2 5
Test for overall effect: Z = 0.00 (P =	1.00)						0.2 0.5 1 2 5 Favours [experimental] Favours [control]
4							ravous (experimental) ravous (control)

#### Forest plot Color Change

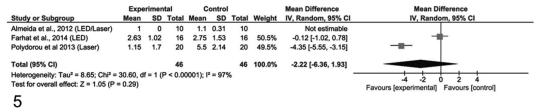


Figure 3. Forest plot for the event "sensitivity intensity."

Figure 4. Forest plot for the event "sensitivity incidence."

Figure 5. Forest plot for the event "color change."

office dental bleaching. Therefore, the initial hypothesis of the study was rejected, which is in agreement with the findings of previous studies. <sup>22,26,27</sup>

Sensitivity after in-office bleaching procedures is directly related to the type and concentration of the bleaching agent, contact time of the bleaching agent with the tooth structure, and photothermal effect of the light source used in a way that is compatible with the application of the bleaching gel. Tooth sensitivity was assessed after bleaching treatment in most of the studies included in the present review. <sup>22-24,27</sup>

Although the literature reports that laser has antiinflammatory and analgesic effects, <sup>28</sup> no reduction in sensitivity was found when this light source was used. This result may be explained by the fact that the laser does not come into direct contact with the pulp. Moreover, the layer formed by the bleaching gel, which has pigments, can minimize the energy density on the target tissue and cause the reflection of light, which significantly reduces the absorption of the laser by the dental pulp. This means that the therapeutic action of the laser may be reduced, resulting in no positive effect on tooth sensitivity. 9,27,29,30

Since no beneficial effects on tooth sensitivity have been found, the use of light sources during the whitening process should be viewed with caution. These sources have a photothermal effect on the bleaching agent, and the conversion of light energy into heat can lead to an uncontrolled increase in intrapulpal temperature. According to Lima and others, a 5°C increase in temperature is harmful to the pulp.

Another possible explanation for the lack of a significant difference in sensitivity (both intensity and incidence) after bleaching with the use of laser compared with other light sources is that most of the studies evaluated 22,24,27 used a hybrid device that combined three blue LED outputs and three low-level infrared laser outputs. This hybridization of light sources can prevent the laser light from being collimated, so that it does not reach the target tissue adequately to provide any beneficial effects regarding a reduction in tooth sensitivity.<sup>27</sup>

Another benefit often expected when using light sources during bleaching is the enhancement of the change in tooth color. Therefore, this was an outcome evaluated in the present review. Despite being widely used,<sup>23</sup> a subjective color analysis is complex because the distinction of color can vary from individual to individual and is influenced by both environmental and physical factors, such as the size, shape, and position of the teeth; ambient light; and background color.<sup>1,5</sup> In the clinical studies included in the meta-analysis for this outcome,<sup>25-27</sup> the results were based on a subjective assessment using a color scale.

No statistically significant difference in the subjective color assessment was found when the use of the laser was compared to LED or halogen light. This result is in agreement with data reported in recent studies<sup>2,3,9,15,32</sup> in which light activation during in-office bleaching was found to exert no influence on the change in tooth color. As the change in color is directly related to the bleaching agent used, its concentration, and contact time with the dental surface, this may explain the lack of significant differences among the light sources evaluated.<sup>2,3</sup>

Although the influence of laser and other light sources is being investigated throughout the world, several characteristics of the studies limit the discussion. In addition, these results should be interpreted with caution given that for the four studies included in the meta-analysis the mean scores for pain in the control group were low. Having low values in the control group may make it difficult to resolve the effect of the treatment studied. Although all studies included in the present systematic review were randomized clinical trials, the large diversity of protocols regarding the application of the

light source and whitening gel as well as the concentration of these agents prevent the direct comparison of results. Further studies with standardized protocols are required as well as studies that use precise, objective color and sensitivity assessment methods to reduce the potential influence of bias.

#### **CONCLUSIONS**

Within the limitations of this study, the evidence in this systematic review and meta-analysis suggests that laser exerts no influence on tooth sensitivity compared with other light sources when used during in-office dental bleaching. The included studies demonstrated that laser use during in-office bleaching may have no influence on the degree of tooth color change.

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#### Conflict of interest

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Clinical Efficiency of Self-etching One-Step and Two-Step Adhesives in NCCL: A Systematic Review and Meta-analysis

CPP de Assis • CAA Lemos • JML Gomes • BCE Vasconcelos SLD Moraes • R Braz • EP Pellizzer

#### Clinical Relevance

One-step self-etch adhesive systems provide a clinical time gain, decreasing the number of clinical steps. When a clinician is able to follow a simpler process of adhesion there is less chance of adhesive failure.

#### **SUMMARY**

Objective: A systematic review and meta-analyses were performed to evaluate whether one-step self-etching (1SSE) adhesive systems are as effective as two-step self-etching (2SSE) adhesives in noncarious cervical lesion (NCCL) restorations.

Methods: This systematic review was conducted according to the guidelines of the Preferred Reporting Items for Systematic Reviews and

\*Cláudio Paulo Pereira de Assis, DDS, MS, PhD candidate, Department of Restorative Dentistry, University of Pernambuco, Camaragibe, Pernambuco, Brazil

Cleidiel Aparecido Araujo Lemos, DDS, MS, PhD, adjunct professor, Department of Dentistry, Federal University of Juiz de Fora (UFJF), Governador Valadares – Minas Gerais, Brazil

Jéssica Marcela de Luna Gomes, DDS, MS, PhD candidate, Department of Dental Materials and Prosthodontics, Araçatuba Dental School, São Paulo State University, Araçatuba, São Paulo, Brazil

Belmiro Cavalcanti do Egito Vasconcelos, PhD, School of Dentistry, Pernambuco University, Camaragibe, Pernambuco, Brazil Meta-analyses (PRISMA) and recorded in the PROSPERO (CRD42018096747). Electronic systematic searches were conducted in the following databases: PubMed/MEDLINE, Scopus, and Cochrane Library for published articles. Only randomized clinical trials that compared 1SSE with 2SSE adhesives systems were selected. The outcomes were retention, postoperative sensitivity, secondary caries, color match, marginal discoloration, marginal adaptation, and anatomical form.

Sandra Lúcia Dantas de Moraes, PhD, School of Dentistry, Pernambuco University, Camaragibe, Pernambuco, Brazil

Rodivan Braz, DDS, MS, PhD, Department of Restorative Dentistry, University of Pernambuco, Camaragibe, Pernambuco, Brazil

Eduardo Piza Pellizzer, PhD, Dental Materials and Prosthodontics, São Paulo State University (Unesp), Araçatuba, São Paulo, Brazil

\*Corresponding author: Avenida General Newton Cavalcanti, 1650, Camaragibe, Pernambuco 54.753-220, Brazil; e-mail: cppassis@gmail.com

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Results: The searches resulted in 476 studies. After applying the eligibility criteria, five randomized controlled trials were selected in which 822 restorations in NCCLs were distributed in 237 patients. The results showed no statistical difference between 1SSE and 2SSE in relation to retention (p=0.23; relative risk [RR]=1.55; 95% confidence interval [CI]=0.76, 3.19), postoperative sensitivity (p=0.50; RR=3.00; 95% CI=0.13, 70.64), Secondary caries (p=0.63; RR=0.68; 95% CI=0.14, 3.31), colormatch (p=0.41; RR=0.64; 95% CI=0.23, 1.83), marginal discoloration (p=0.93; RR=1.02; 95% CI=0.65, 1.61), and anatomical form (p=0.56: RR=1.38; 95% CI=0.46, 4.13). However there was statistical difference in relation to marginal adaptation (p=0.01; RR=1.95; 95% CI=1.14, 3.34).

Conclusion: This systematic review with metaanalysis revealed that both 1SSE and 2SSE adhesive systems have comparable clinical effectiveness in a follow-up period of 12 to 24 months, except in relation to marginal adaptation.

#### INTRODUCTION

Adhesive systems have undergone important changes in recent years, mainly by simplifying their application, without compromising adhesion to the dental substrates. <sup>1,2</sup> At first, three-step etch-andrinse adhesives systems were available, and soon after, two-step self-etching (2SSE) systems were introduced. Now, one-step universal adhesive systems are available. <sup>1</sup>

One-step self-etching (1SSE) adhesives provide easy clinical application, reduce technical sensitivity, and are well accepted by clinicians. Although 1SSE adhesives have a simplified approach, early formulations did not promote effective dentin sealing. However, manufacturers have modified the chemical formulations of new one-step adhesives to improve their clinical performance.<sup>3,4</sup>

Two-step adhesives consist of acidic monomers dissolved in aqueous solution and a layer of hydrophobic resin as a second step. Single-step adhesives do not have this hydrophobic layer. The degree of demineralization of acidic monomers in self-etching adhesives depends on their pH, which may be mild, moderate, or strong. Self-etching adhesives are able to infiltrate the smear layer and partially dissolve the hydroxyapatite, generating a hybrid layer with incorporated minerals. The current trend is to use

simplified adhesive materials, which are available from many manufacturers. Self-etching adhesive systems have become popular for clinicians<sup>4</sup> because they do not require preconditioning with phosphoric acid or an overwashing step; they also provide a clinical time gain over etch-and-rinse adhesives.

Generally, noncarious cervical lesions (NCCLs) are used as determinants of the clinical effectiveness of adhesives. This type of restoration is usually caused by stress in the cervical region of the teeth, and the cavity formed involves dentin, which makes adhesion more difficult. In addition, NCCLs present high prevalence and easy access to restoration (located in the vestibular region), do not require complicated restorative techniques, can be considered free cavities because they have a low polymerization contraction factor, and do not usually provide macromechanical retention. 3,6

Currently, there is no consensus in the literature regarding the best adhesive indicated for clinical use in the restoration of NCCLs. The aim of this systematic review with meta-analyses was to evaluate whether 1SSE adhesive systems are as effective as 2SSE adhesives in NCCLs. The null hypothesis was that there is no difference between 1SSE and 2SSE adhesive systems for restoration retention. The second hypothesis was that there is no difference between 1SSE and 2SSE adhesives for postoperative sensitivity, secondary caries, color match, marginal discoloration, marginal adaptation, and anatomical form.

#### **METHODS AND MATERIALS**

#### **Registration Protocol**

This systematic review was conducted according to the guidelines of the Preferred Reporting Items for Systematic Reviews and Meta-analyzes (PRISMA) checklist.<sup>8</sup> This systematic review was recorded in the International Prospective Register of Systematic Reviews (PROSPERO) under registration number CRD42018096747.

#### **Eligibility Criteria**

The investigated question of this study was "Are there any differences in outcomes between the 1SSE and 2SSE adhesive systems?" based on PICO criteria. In view of this, the population (P) consisted of patients with restorations in which self-etching adhesive systems were used; the intervention (I) consisted of patients with restorations using 1SSE adhesive; comparison (C) was patients with restorations using 2SSE adhesive; and outcomes (O) evaluated were

retention of restoration (primary outcome) and postoperative sensitivity, secondary caries, color match, marginal discoloration, marginal adaptation, and anatomical form (secondary outcomes).

The inclusion criteria used were randomized clinical trials (RCTs), studies with a minimum follow-up of 12 months, studies evaluating direct restorations in NCCLs of permanent teeth using 1SSE and 2SSE adhesives. The exclusion criteria were nonrandomized and retrospective studies, case reports, reviews, *in vitro* studies, animal studies, computer simulations, studies that evaluated selfetch adhesive systems with etch-and-rinse adhesives, studies reported in more than one publication with different follow-up periods and published report reviews.

#### **Information Sources and Search Strategy**

The electronic search of the literature was performed by two reviewers (CPPA and CAAL) working independently. Studies were selected and included/ excluded based on the article title and abstract in the PubMed/MEDLINE, Scopus, and Cochrane Library databases using the key words "Adhesive dental AND one-step AND two-step". To complement this review, the same researchers conducted a manual search for articles published in the following journals: Operative Dentistry, Dental Materials, Journal of Dentistry, Journal of Adhesive Dentistry, American Journal of Dentistry, Brazilian Dental Journal, and Clinical Oral Investigations. In addition, Open-Grey (www.opengrey.eu) was used to search gray literature. The electronic search was conducted until July 2019 without limiting the year of publication during searches.

The studies were initially selected and classified according to the inclusion/exclusion criteria by reading the title and abstract. Studies that did not clearly fit the inclusion/exclusion criteria were downloaded and read in full, and then a decision was made to include or exclude in the review. A third researcher (EPP) analyzed all the differences in choices between the researchers, and a consensus was achieved.

#### **Data Analysis**

One of the authors (CPPA) collected important information from the articles, and a second author (CAAL) reviewed all the information collected. A careful analysis was performed to verify disagreements between the authors, and a third author (EPP) was consulted to obtain consensus.

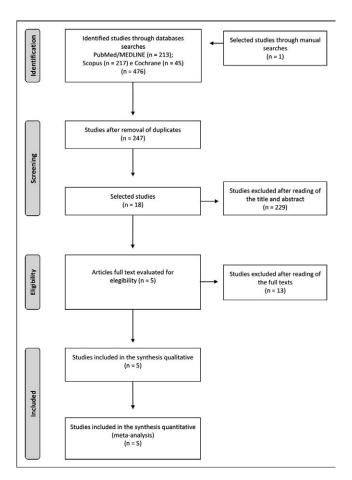


Figure 1 Flow diagram of the study.

#### Risk of Bias

Two investigators (CPPA and CAAL) evaluated the methodologic quality of the studies according to the Cochrane bias risk tool for RCT studies to verify the level of evidence from the studies included in the review.

#### **Summary Measures**

The meta-analyses were based on the Mantel-Haenzel and inverse variance methods. 1SSE and 2SSE were used in the study to assess the effects of the treatment on the body. The relative risk (RR) and 95% confidence interval (CI) were calculated for each study, The RR values were considered significant at p < 0.05. The extracted data were analyzed using Review Manager software (RevMan) 5.3 (The Cochrane Collaboration, Copenhagen, Denmark).

#### **Additional Analysis**

The Kappa score<sup>9</sup> test was used to calculate the level of concordance between authors during the article selection process in the PubMed/MEDLINE, Scopus,

Table 1: Reasons Studies Were Excluded (n=13)								
Reason								
Only two-step self-etching adhesive system <sup>1</sup>								
Only one-step self-etching adhesive system <sup>10</sup>								
Class I and II restorations <sup>11</sup>								
Orthodontic bracket adhesion failure <sup>12</sup>								
Etch-and-rinse adhesive system <sup>13-19</sup>								
Sealant evaluation <sup>20</sup>								
Primary teeth <sup>21</sup>								

and Cochrane Library databases. Any disagreements were resolved by discussion and the consensus of all authors.

#### **RESULTS**

#### Literature Search

The initial database search resulted in 476 references, 213 in PubMed/MEDLINE, 217 in Scopus, 45 in the Cochrane Library, and 1 via manual searches. After duplicate references were removed, a detailed review of the titles and abstracts of the selected studies was performed, and 247 articles remained. After detailed analysis, the inclusion/exclusion criteria were applied, and 18 complete articles were downloaded and selected for further analysis. After reading the articles completely, 13 were excluded (Table 1), resulting in five articles selected for this systematic review and meta-analysis (Figure 1). The level of agreement among researchers during the initial article selection process was PubMed/MED-LINE (0.91), Scopus (0.90), and the Cochrane Library (1.00). The values indicated a high level of agreement among the reviewers according to the Kappa criterion.

#### **Characteristics of Included Studies**

The characteristics of the included studies are detailed in Table 2. All five articles selected were RCTs.<sup>7,22-25</sup> In total, 822 NCCL-type restorations were performed in 237 patients with a mean age of

45 years. Four different 1SSE adhesives systems and three 2SSE adhesive systems were used (Table 2). The mean follow-up was 18 months.

The main inclusion criteria for the studies were NCCLs with no more than three restorations per study participant. <sup>22</sup> We found that 46.4% of the restorations were maxillary and 53.6% were mandibular, with a fairly homogeneous distribution of restorations. In general, restorations did not involve more than 50% of the cavosurface margin in enamel, and 75% of the restoration surface was in dentin.

In all five included studies, <sup>7,22-25</sup> researchers evaluated their outcomes through the United States Public Health Service criteria, and usually the follow-up examinations were performed every three months with a maximum follow-up of 24 months. All five studies evaluated retention of the restoration, besides marginal discoloration, secondary caries, and marginal adaptation. Only three studies assessed anatomical form<sup>22,23,25</sup> and postoperative sensitivity. <sup>7,22,24</sup> Among the included studies, four<sup>22-25</sup> evaluated color match. Detailed data from the studies are described in Table 3.

The one-step adhesive systems used were Xeno III (Dentsply, York, PA, USA), Clearfil S3 Bond (Kuraray, Okayama, Japan), Adper Easy Bond (3M ESPE, St Paul, MN, USA), Xeno V + (Dentsply), and the two-step systems were Clearfil Protect Bond (Kuraray); Clearfil SE Bond (Kuraray), and Adper Scotchbond SE (3M ESPE).

Three composite resins were used. One study  $^{23}$  used Filtek Supreme Plus (3M ESPE), two studies  $^{7,22}$  used Esthet-X HD (Dentsply), and two other studies  $^{24,25}$  used AP-X resin, (Kuraray).

#### Assessment of the Risk of Bias

The risk of bias was assessed using the Cochrane bias risk tool for RCT studies to verify the level of evidence from the studies. The findings indicated a high risk of bias for blinding of participants<sup>22,23</sup>; an unclear risk of bias to allocation, blinding of

Author, Year,	Study	Patients,	NCCL,	Average	Follow-up,	Adhesi	ve System	
Reference		n	n	Age, y	mo	Intervention: 1SSE	Control: 2SSE	
Pena and others (2016) <sup>7</sup>	RCT	25	112	NR	24	Xeno V+ (n=28)	Clearfil SE Bond (n=28)	
Türkün and others (2005) <sup>22</sup>	RCT	35	163	44	12	Xeno III (n=75)	Clearfil Protect Bond (n=85)	
Perdigão and others (2012) <sup>23</sup>	RCT	39	125	47.6	18	Adper Easy Bond (n=28)	Adper Scotchbond SE (n=26)	
Zhou and others (2009) <sup>24</sup>	RCT	124	342	42.5	12	Clearfil Tri-S Bond (n=115)	Clearfil SE Bond (n=116)	
Brackett and others (2010) <sup>25</sup>	RCT	14	80	46	24	Clearfil S3 Bond (n=40)	Clearfil SE Bond (n=40)	
Abbreviations: 1SSE, one-step self-etching; 2SSE, two-step self-etching; NCCL, noncarious cervical lesion; NR, not reported; RCT, randomized clinical trial.								

Author, Year, Reference	1SSE		2SSE	
	Failure	Total	Failure	Tota
Pena and others (2016) <sup>7</sup>	Retention (n=1)	28	Retention (n=0)	28
	Postoperative sensitivity (n=1)	-	Postoperative sensitivity (n=0)	=
	Secondary caries (n=0)	-	Secondary caries (n=0)	_
	Color match (non)	-	Color match (non)	_
	Marginal discoloration (n=5)	•	Marginal discoloration (n=1)	=
	Marginal adaptation (n=2)	-	Marginal adaptation (n=0)	=
	Anatomical form (non)	-	Anatomical form (non)	_
Türkün and others (2005) <sup>22</sup>	Retention (n=3)	75	Retention (n=0)	85
	Postoperative sensitivity (n=0)	•	Postoperative sensitivity (n=0)	='
	Secondary caries (n=0)	-	Secondary caries (n=0)	_
	Color match (n=0)	-	Color match (n=1)	_
	Marginal discoloration (n=2)	-	Marginal discoloration (n=1)	_
	Marginal adaptation (n=1)	-	Marginal adaptation (n=0)	_
	Anatomical form (n=2)	•	Anatomical form (n=1)	_
Perdigão and others (2012) <sup>23</sup>	Retention (n=2)	26	Retention (n=2)	22
	Postoperative sensitivity (n=0)	_	Postoperative sensitivity (n n=0)	
	Secondary caries (n=2)	-	Secondary caries (n=2)	_
	Color match (n=2)	_	Color match (n=3)	
	Marginal discoloration (n=8)	_	Marginal discoloration (n=8)	
	Marginal adaptation (n=14)	_	Marginal adaptation (n=6)	
	Anatomical form (n=4)		Anatomical form (n=3)	
Zhou and others (2009) <sup>24</sup>	Retention (n=3)	115	Retention (n=2)	116
	Postoperative sensitivity (n=0)		Postoperative sensitivity (n=0)	_
	Secondary caries (n=0)	_	Secondary caries (n=0)	
	Color match (n=1)	_	Color match (n=2)	
	Marginal discoloration (n=8)	-	Marginal discoloration (n=8)	_
	Marginal adaptation (n=4)	_	Marginal adaptation (n=3)	
	Anatomical form (none)		Anatomical form (none)	
Brackett and others (2010) <sup>25</sup>	Retention (n=7)	37	Retention (n=6)	37
	Postoperative sensitivity (n=none)	_	Postoperative sensitivity (n=none)	<u>-</u>
	Secondary caries (n=0)	_	Secondary caries (n=1)	_
	Color match (n=2)	_	Color match (n=2)	<u></u>
	Marginal discoloration (n=8)	_	Marginal discoloration (n=12)	_
	Marginal adaptation (n=4)	_	Marginal adaptation (n=6)	_
-	Anatomical form (n=6)		Anatomical form (n=1)	

participants,<sup>7,24</sup> and incomplete outcome<sup>25</sup>; and a low risk for other biases, where it was shown that the studies were of high quality (Figure 2).

#### **Meta-analyses**

*Primary Outcome*—Five studies<sup>7,22-25</sup> were selected for quantitative analysis comparing 1SSE adhesive systems and 2SSE adhesive systems. The meta-analysis showed no statistically significant differ-

ence between 1SSE and 2SSE regarding retention (p=0.23; RR=1.55; 95% CI=0.76, 3.19) (Figure 3).

Secondary Outcome—Regarding postoperative sensitivity, three studies  $^{7,22,24}$  were included for quantitative analysis. The data showed no statistically significant difference between 1SSE and 2SSE (p=0.50; RR=3.00; 95% CI=0.13, 70.64) (Figure 4). The same was observed for secondary caries  $^{7,22-25}$  (p=0.63; RR=0.68; 95% CI=0.14, 3.31) (Figure 5), color match  $^{22-25}$  (p=0.41; RR=0.64; 95% CI=0.23,

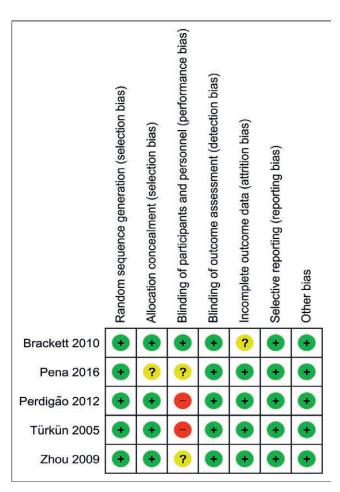


Figure 2. Risk of study bias.

1.83) (Figure 6), marginal discoloration<sup>7,22-25</sup> (p=0.93; RR=1.02; 95% CI=0.65, 1.61) (Figure 7), and anatomical form<sup>22-25</sup> (p=0.56; RR=1.38; 95% CI=0.46, 4.13) (Figure 8). However, there was statistical difference in relation to marginal adaptation favorable to group 2SSE (p=0.01; RR=1.95; 95% CI=1.14, 3.34) (Figure 9).

#### **DISCUSSION**

The null hypothesis of this study stating that there is no difference between 1SSE and 2SSE adhesives regarding restoration retention systems was accepted. The meta-analysis showed that there was no statistically significant difference between the results for 1SSE and 2SSE (p=0.23; RR=1.55; 95% CI=0.76, 3.19) (Figure 3). These data are in agreement with other studies. <sup>24,26,27</sup> It is worth mentioning that the similarity between self-etching adhesive systems will allow a greater use of 1SSE systems since they will promote simplification in the technique, optimizing clinical time. <sup>3</sup>

All restorations evaluated in the included studies<sup>7,22-25</sup> were performed in NCCLs because clinical evaluation of adhesive systems is often performed in this type of cavity.<sup>5</sup> They present a greater amount of sclerosed dentin, occlusal forces that emphasize the cervical third of teeth, minimal retention, and margins that are not only enamel but also extend to the dentin<sup>4</sup>; they are also subject to high stress during chewing 28,29 and are anatomically located in a region where there is a high dissipation of occlusal forces, factors that contribute to difficulty in retention. However, all the restorations evaluated presented clinical requirements proposed by the American Dental Association requiring a survival rate ≥90% of the restorations placed after 18 months of follow-up, regardless of the type of adhesive system (1SSE or 2SSE).<sup>7</sup>

Pena and others<sup>7</sup> state that there is a similarity between systems only initially to restorative treatment and that with increasing follow-up, one-step systems are not able to support mechanical fatigue or hydrolysis, causing failure. This is related to the application technique of adhesive systems. In twostep adhesive systems (2SSE), the primer acid is applied, with subsequent application of hydrophobic monomers. <sup>7,30,31</sup> In the one-step system (1SSE), the composition of hydrophilic and hydrophobic monomers are present in the same solution. Hydrophilic monomers can collect water on the substrate or adhesive. 1 The clinician can control this mechanism of water sorption at the moment the adhesive is applied through the correct drying of the substrate, thereby avoiding risks of failure. However, when evaluating the data from the included studies, it was not possible to observe a correlation between the one-step technique and the failures of the restorations.

A systematic review<sup>3</sup> found an increase in postoperative sensitivity when using conventional adhesive systems compared with self-etching adhesive systems. Also, among the self-etching adhesives, some studies<sup>7,23</sup> stated that one-step adhesives (1SSE) allowed a greater control of postoperative sensitivity. However, the included studies found similarity between the systems (1SSE and 2SSE) (p=0.50; RR=3.00; 95% CI=0.13, 70.64), which is justified by the presence of dentin sclerosis in NCCLs and composite resin that will serve as insulators, thus masking the role of the adhesive system in the postoperative sensitivity.

The meta-analyses showed no difference between the 1SSE and 2SSE adhesive systems for postoperative sensitivity, occurrence of secondary caries,

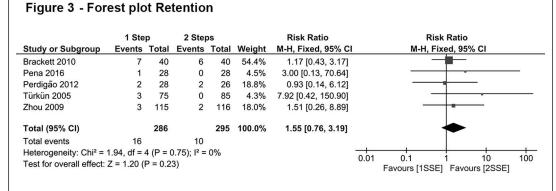


Figure 4 - Forest plot Postoperative Sensitivity

	1 Ste	р	2 Step	os		Risk Ratio	Risk Ratio
Study or Subgroup	<b>Events</b>	Total	<b>Events</b>	Total	Weight	M-H, Fixed, 95% CI	M-H, Fixed, 95% CI
Pena 2016	1	28	0	28	100.0%	3.00 [0.13, 70.64]	<del></del>
Türkün 2005	0	75	0	85		Not estimable	
Zhou 2009	0	115	0	116		Not estimable	
Total (95% CI)		218		229	100.0%	3.00 [0.13, 70.64]	
Total events	1		0				
Heterogeneity: Not ap	plicable						0.001 0.1 1 10 1000
Test for overall effect:	Z = 0.68 (	P = 0.5	0)				Favours [1SSF] Favours [2SSF]

Figure 5 - Forest plot Secondary Caries

	1 Ste	р	2 Step	os		Risk Ratio	Risk Ratio
Study or Subgroup	Events	Total	<b>Events</b>	Total	Weight	M-H, Fixed, 95% CI	M-H, Fixed, 95% CI
Brackett 2010	0	40	1	40	42.0%	0.33 [0.01, 7.95]	<del></del>
Pena 2016	0	28	0	28		Not estimable	
Perdigão 2012	2	28	2	26	58.0%	0.93 [0.14, 6.12]	<del></del>
Türkün 2005	0	75	0	85		Not estimable	
Zhou 2009	0	115	0	116		Not estimable	
Total (95% CI)		286		295	100.0%	0.68 [0.14, 3.31]	
Total events	2		3				
Heterogeneity: Chi2 = 0	0.30, df =	1 (P = 0	0.58); I <sup>2</sup> =	0%			004 04 4 40 400
Test for overall effect:	Z = 0.48 (	P = 0.6	3)				0.01 0.1 1 10 100 Favours [1SSE] Favours [2SSE]

Figure 6 - Forest plot Color Match

	1 Step		2 Steps			Risk Ratio	Risk Ratio
Study or Subgroup	Events	Total	<b>Events</b>	Total	Weight	M-H, Fixed, 95% CI	M-H, Fixed, 95% CI
Brackett 2010	2	40	2	40	23.5%	1.00 [0.15, 6.76]	<del></del>
Perdigão 2012	2	28	3	26	36.6%	0.62 [0.11, 3.41]	<del></del>
Türkün 2005	0	75	1	85	16.5%	0.38 [0.02, 9.12]	-
Zhou 2009	1	115	2	116	23.4%	0.50 [0.05, 5.49]	-
Total (95% CI)		258		267	100.0%	0.64 [0.23, 1.83]	
Total events	5		8				
Heterogeneity: Chi² = 0.35, df = 3 (P = 0.95); l² = 0%							
Test for overall effect: Z = 0.83 (P = 0.41)						0.01	

Figure 3. Forest plots of the retention outcome.

Figure 4. Forest plots of the postoperative sensitivity outcome.

Figure 5. Forest plots of the secondary caries outcome.

Figure 6. Forest plots of the color match outcome.

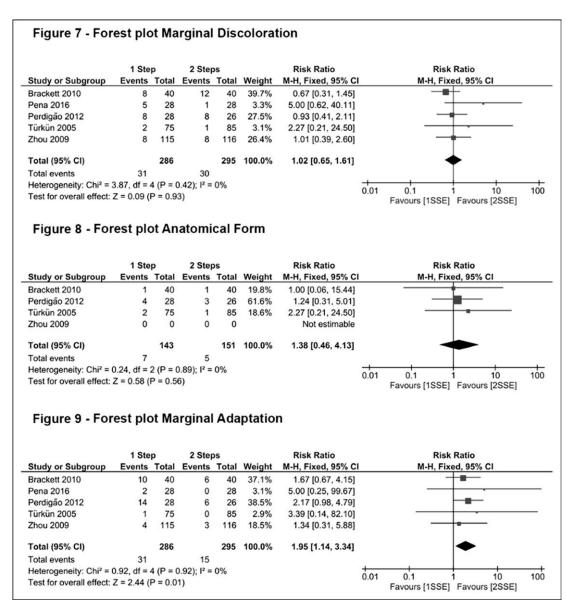


Figure 7. Forest plots of the marginal discoloration outcome.

Figure 8. Forest plots of the anatomical form outcome.

Figure 9. Forest plots of the marginal adaptation outcome.

color match, marginal discoloration, and anatomical form. However, regarding marginal adaptation, there was statistical difference favorable to group 2SSE (p=0.01; RR=1.95; 95% CI=1.14, 3.34), so the second hypothesis of this study was rejected. One of the difficulties of adhesion on the margins of NCCLs is that they have a substrate in both dentin and enamel.<sup>32</sup> Studies<sup>23,26</sup> have found that a disadvantage of the use of self-etching adhesives (1SSE and/or 2SSE) is in deficient acid conditioning of enamel compared with conventional systems.

However, the results of this systematic review and meta-analysis, which only looked at self-etching adhesive systems (1SSE and 2SSE), showed minimal adjustments regarding retention, color match, marginal discoloration and anatomical form in enamel<sup>7,25</sup> and excellent results of absence of secondary caries. Similar results were found in other studies.<sup>4,32</sup>

All included studies were RCTs; clinical trials can provide reliable and direct evidence to guide clinicians in choosing dental materials, RCTs represent

the standard design for evaluating health care interventions. Well-designed RCTs and systematic reviews of well-designed RCTs are on the top of the hierarchy of the levels of evidence. The quality of the studies was analyzed from the Cochrane scale, where the high risk of bias observed for blinding is justified by the clinical technique used in applying the adhesive, which makes it difficult to screen the examiners. The results of this review should be interpreted with caution because of the small number of clinical trials evaluated. Other RCTs with longer observation periods are still needed.

#### **CONCLUSIONS**

This systematic review with meta-analysis revealed that both 1SSE and 2SSE adhesive systems have comparable clinical effectiveness in a follow-up period of 12 to 24 months, except for marginal adaptation.

#### **Acknowledgements**

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#### **Conflict of Interest**

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Effect of Deep Margin Elevation on CAD/CAM-Fabricated Ceramic Inlays

TJ Vertolli • BD Martinsen • CM Hanson • RS Howard • S Kooistra • L Ye

#### Clinical Relevance

Using the deep margin elevation technique in preparations extending beyond the cementoenamel junction appears to be beneficial in maintaining structural integrity of CAD/CAMfabricated feldspathic ceramic inlays.

#### **SUMMARY**

Objective: To evaluate the effect of deep margin elevation on structural and marginal integrity of ceramic inlays.

Methods and Materials: Forty extracted human third molars were collected and randomly separated into four groups (n=10/group). In group 1 (enamel margin group), the gingival margin was placed 1 mm supragingival to the cemento-enamel junction (CEJ). In group 2 (cementum margin group), the gingival margin

Travis J. Vertolli, DMD, MS, Comprehensive Dentistry resident, Naval Postgraduate Dental School, Navy Medicine Professional Development Center, and Postgraduate Dental College, Uniformed Services University of Health Sciences, Bethesda, MD, USA

Bradley D. Martinsen, DMD, MS, Comprehensive Dentistry resident, Naval Postgraduate Dental School, Navy Medicine Professional Development Center, and Postgraduate Dental College, Uniformed Services University of Health Sciences, Bethesda, MD, USA

Christopher M. Hanson, DDS, MS, Comprehensive Dentistry resident, Naval Postgraduate Dental School, Navy Medicine Professional Development Center, and Postgraduate Dental College, Uniformed Services University of Health Sciences, Bethesda, MD, USA

Robin S. Howard, MA, supervisory biostatistician, assistant chief, Department of Research Programs, Walter Reed National Military Medical Center, Bethesda, MD, USA was placed 2 mm below the CEJ. In group 3 (glass ionomer [GI] margin group), the gingival margin was placed 2 mm below the CEJ, and then the margin elevated with GI to the CEJ. In group 4 (resin-modified glass ionomer [RMGI] margin group), the gingival margin was placed 2 mm below the CEJ, and then the margin elevated with RMGI to the CEJ. Standardized ceramic class II inlays were fabricated with computer-aided design/computer-aided manufacturing and bonded to all teeth, and ceramic proximal box heights were measured. All teeth were subjected to 10,000 cycles of thermocycling (5°C/55°C) and then underwent

Scott Kooistra, DDS, MS, professor, chairman, Operative Dentistry Department, Naval Postgraduate Dental School, Navy Medicine Professional Development Center, and Postgraduate Dental College, Uniformed Services University of Health Sciences, Bethesda, MD, USA

\*Ling Ye, DDS, PhD, associate professor, chairman, Research Department, Naval Postgraduate Dental School, Navy Medicine Professional Development Center, and Postgraduate Dental College, Uniformed Services University of Health Sciences, Bethesda, MD, USA

\*Corresponding author: 8955 Wood Rd, Building One, Bethesda, MD 20889-5628, USA; e-mail: ling.ye.mil@mail.mil

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1,200,000 cycles of vertical chewing simulation at 50 N of force. Ceramic restorations and marginal integrity were assessed with a Hirox digital microscope. The Fisher exact test (two-tailed) with adjusted p-values ( $\alpha$ =0.05) and logistic regression were used for statistical analysis.

Results: The cementum margin group had a significantly higher ceramic fracture rate (90%) compared to other groups (10% in enamel margin and GI margin groups, p=0.007; 0% in RMGI group, p<0.001). Logistic regression showed that with increased ceramic proximal box heights, the probability of ceramic fracture increased dramatically.

Conclusion: Deep marginal elevation resulted in decreased ceramic fracture when preparation margins were located below the CEJ. There was no difference found between margin elevation with GI or RMGI. Increased heights of ceramic proximal box may lead to an increased probability of ceramic fracture.

#### INTRODUCTION

In today's society, patients' desires as well as advances in computer technologies have drastically changed the dental treatment landscape. Public demand has forced today's dental treatment to become more esthetic and more immediate. Traditionally, large, deep carious lesions in posterior teeth were restored with amalgam or indirect cast gold restorations. The use of amalgams has been drastically decreased. 1,2 The trend toward nonamalgam restorations has been enhanced by teaching the use of amalgam alternatives in many dental schools.3 The indirect all-ceramic restoration has been developed as an esthetic alternative to amalgam, gold, and metal-ceramic restorations. 4 Additionally, computer-aided design/computer-aided manufacturing (CAD/CAM) advances have given clinicians the ability to create definitive indirect ceramic restorations in one visit, appeasing the patient's desire for an immediate return on investment. CAD/CAM eliminates the need for traditional impressions, stone casts, and, sometimes, provisional restorations.<sup>5</sup> CAD/CAM-fabricated restorations have shown to be reliable up to 18 years with marginal integrity similar to crowns fabricated by traditional laboratory methods.<sup>3,6</sup> Advances in resin bonding techniques have helped all-ceramic restorations become extremely retentive to tooth structure, especially when margins can be placed on enamel.<sup>7</sup> Resin bonding has also been shown in laboratory studies to significantly increase the flexural strength of many all-ceramic crown materials.<sup>8</sup>

Yet, even with these advances in material, design, and manufacturing sciences, deep subgingival carious lesions or deep defective restorations remain a significant restorative challenge. The ideal margin location for an all-ceramic restoration is one with adequate enamel available to bond with resin cement.9 As carious lesions and restorations become larger and deeper, gingival marginal enamel thins out until reaching the cemento-enamel junction (CEJ), at which point bonding to enamel is no longer possible. Margins placed apical to the CEJ on dentin are more prone to microleakage, which is caused by both shrinkage during curing and differences in the coefficient of thermal expansion between restorative material and tooth structure 10 as well as incomplete hybridization between adhesive system and the collagen fibrils due to entrapped water between interfibrillar spaces. 11 Microleakage may lead to secondary caries and eventual restoration failure. 12,13 Resin bonding is more technique sensitive, 2 and the apical margin of the restoration should be placed in enamel if possible. 14 Gargiulo and others 15 have described the soft tissue attachment coronal to crestal bone as the biologic width, made up of connective and epithelial attachment. Placing restoration margins that invade the biologic width, generally taken to be 3 mm coronal to the bony crest, can cause gingival inflammation, loss of periodontal attachment, and bone resorption. <sup>16</sup> Deep subgingival margins may encroach on or invade the biologic width. On the other hand, supragingival margins make impression making, whether digital or traditional, easier and more accurate. When bonding indirect ceramic restorations, removing excess cement and polishing margins are much easier to accomplish if margins are located in a supragingival position.<sup>17</sup>

Several treatment options allow these deep restorative margins to be placed in a more manageable, supragingival position. Orthodontic extrusion is one option but can take months and can result in esthetic compromise due to root form and difficulty creating a natural emergence profile. Surgical crown lengthening can also give better access to deep margins. However, after healing from crown lengthening, esthetic compromise and root hypersensitivity are possible complications. To Crown lengthening compromises adjacent alveolar bone support and may affect future implant therapy. While the traditional crown-lengthening procedure still has its place in dentistry, newer, minimally invasive procedures

have been demonstrated to show success even with restorative margins encroaching on biologic width, as demonstrated in the article by Sarfati and Tirlet<sup>18</sup> evaluating three clinical cases where deep restoration margins were well tolerated by the surrounding periodontium, clinically and histologically. Other case reports have shown restorations invading biologic width that maintained a periodontium free of gingival and periodontal inflammation as long as they had smooth, well-contoured margins along with meticulous oral hygiene maintenance by the patient. <sup>16</sup>

The third option—and the focus of this study—is deep margin elevation (DME). Also known as proximal box elevation or cervical margin relocation, this nonsurgical technique uses a direct restoration placed only at the deep apical portion of the preparation to elevate the margin to a more coronal and more conducive position for final restoration fabrication and cementation.<sup>19</sup> Also referred to as the open sandwich technique, DME leaves the direct restoration exposed to the oral environment. This additional interface of direct restoration has the potential for leakage, and there are concerns that an increased failure rate may be associated with this technique. While a lot of literature has used resin composite as the direct restorative material to margin elevate beneath all-ceramic indirect restorations, 3,7,20,21 some recent literature has advocated the use of glass ionomer (GI) or resin-modified glass ionomer (RMGI) to elevate deep margins. 18,22 Traditional GIs are a mixture of alumino-fluoro-silicate glass particles and polyalkenoic acid. They set as a result of a chemical reaction on mixing that requires water to facilitate ionic exchange.<sup>23</sup> Hence, they perform well in humid environments, such as a deep subgingival preparation in damp, tubular dentin.<sup>24</sup> RMGIs have a photopolymerizable resin in addition to the traditional GI formulation.<sup>25</sup> RMGI has better physical properties, including increased cohesive strength, in addition to the high compressive strength of traditional GI. RMGI also has increased polishability and esthetic results due to decreased filler particle size.<sup>26</sup>

GI/RMGI restorative materials have several material characteristics that would lend them to be potentially a better restorative material for use in DME than traditional or flowable resin composites. First, the coefficient of thermal expansion of GIs is closest to dentinal tissues; thus, thermal stresses over time have less effect on the marginal interface, resulting in less microleakage.<sup>27</sup> Second, as mentioned above, the hydrophilic nature of GIs is better

for bonding in deep dentin, which will be damp due to the amount of dentinal tubules present.<sup>24</sup> Third, in areas with no enamel for resin bonding, GIs form a strong chemical bond to tooth structure via chelation. Ionic bonds form between carboxyl groups of the polyalkenoic acid and the hydroxyapatite.<sup>28</sup> This bond matures over the weeks after placement, increasing in strength. Fourth, GIs have a low modulus of elasticity, a relative "flexibility" that lessens internal stress and stiffness after cure, helping to prevent debonding.<sup>29</sup> The low modulus of elasticity allows the restoration to act as a stressabsorbing layer, relieving contraction stresses and improving marginal integrity.<sup>30</sup> On the other hand, resin composite's high polymerization shrinkage and higher modulus of elasticity increase the likelihood of marginal leakage following curing. 11 Finally, GIs release fluoride and can also be recharged by topical fluoride. 31,32 In deep subgingival areas more prone to secondary caries, fluoride-releasing materials pass fluoride across the marginal gap to the tooth structure, forming fluorapatite.<sup>33</sup> As described by Featherstone,<sup>34</sup> fluoride prevents caries in three ways: inhibiting bacterial metabolism, inhibiting demineralization, and enhancing remineralization—all three important tasks in the subgingival environment. There are several properties of GI/ RMGI that are less ideal when compared to resin composite, including a less polishable surface as well as higher solubility rates. Despite these qualities, GI/RMGI restorations remain clinically acceptable for use.<sup>3</sup>

As demonstrated, DME is a conservative and efficient restorative technique used when restoration margins are deep subgingival, likely beyond the CEJ. While GI restorations seem to have many characteristics that would be beneficial in the subgingival environment, there is little literature regarding GI's use in DME. Therefore, the purpose of this study is to evaluate the effect of DME with GI/RMGI on the structural and marginal integrity of CAD/CAM-fabricated ceramic inlays.

#### **METHODS AND MATERIALS**

#### **Study Design**

This laboratory study assessed both structural and marginal integrity of CAD/CAM-fabricated ceramic inlays. Independent variables consisted of 1) gingival margin position (enamel and cementum) and 2) margin elevation restorative material (GI and RMGI). Dependent variables, or outcomes, were structural and marginal integrity, assessed by visualizing any fracture of the feldspathic ceramic

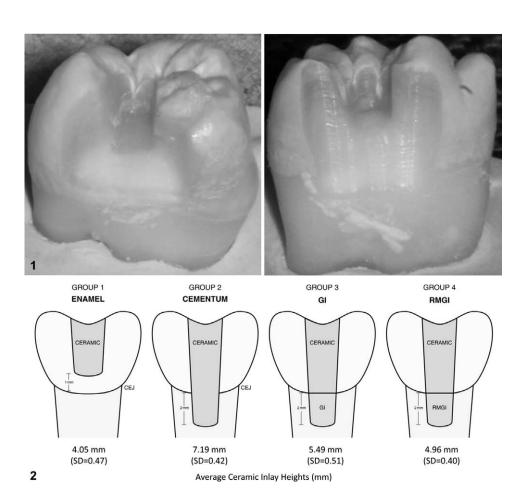


Figure 1. Tooth preparation in the enamel margin group (left): The gingival margin of the preparation was placed 1 mm above the cementoenamel junction (CEJ). Tooth preparation in the cementum margin group (right): The gingival margin of the preparation was placed 2 mm below the CEJ.

Figure 2. Schematic illustration of the four experimental groups: enamel margin group, cementum margin group, glass ionomer (GI) margin group, and resin-modified glass ionomer (RMGI) margin group. For GI/RMGI margin groups, the gingival margin of the preparation was placed 2 mm below the cemento-enamel junction (CEJ), then 2 mm of GI/RMGI was added to the CEJ, respectively. Ceramic proximal box heights were measured and are shown as mean and standard deviation (SD).

inlays or gaps in the marginal resin cement interface. Power analysis was used to determine a sample size of 10 per group and could identify a difference of 20% in the risk of structural and marginal integrity compromise between the experimental groups.

#### **Specimen Preparation**

Forty noncarious, unrestored extracted deidentified human third molars were acquired from the National Institute for Dental and Craniofacial Research (NIDCR). Any remaining biologic debris and potential contaminants were removed, and the teeth were stored in 0.5% chloramine T (Sigma-Aldrich, St Louis, MO, USA) at 4°C. Twenty-four hours prior to tooth preparation, all specimens were transferred to deionized water at 4°C. After preparation and restoration placement, the teeth were maintained in deionized water until they were thermomechanically loaded.

Specimens were placed into one of four treatment groups based on the material on the gingival floor of the preparation adjacent to the ceramic inlay. The four groups were designated as follows: group 1: enamel margin; group 2: cementum margin; group 3: GI margin; and group 4: RMGI margin (n=10 per group). Standardized class II proximal ceramic inlay preparations were made (33% of overall width at the bucco-lingual dimension of isthmus, 33% of overall width at the bucco-lingual dimension of proximal box, and 33% of overall occlusal depth, extended to the central groove mesio-distally and 2 mm mesiodistally of axial depth in the proximal box at the gingival margin). For the enamel margin group, the gingival margin of the preparation was placed 1 mm above the CEJ on the enamel tooth structure. In the remaining three groups, the preparation ended 2 mm below the CEJ in cementum (Figure 1). The 10 teeth in the GI margin group had 2 mm of deep margin elevation to the CEJ with self-cure GI (Fuji IX, GC America, Alsip, IL, USA), and the 10 teeth in the RMGI margin group, had 2 mm of deep margin elevation to the CEJ with dual-cured RMGI (Fuji II LC, GC America).

Following specimen preparation and margin elevation (GI and RMGI groups only), all 40 preparations were scanned with the CEREC Omnicam

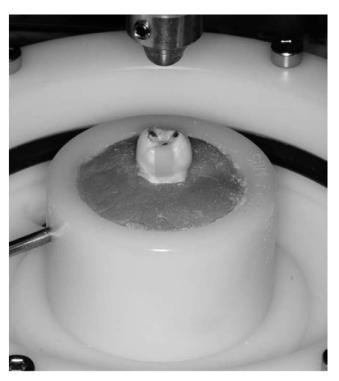


Figure 3. Specimen was mounted in acrylic resin and prepared for chewing simulation.

(Dentsply Sirona, Charlotte, NC, USA). Forty feld-spathic porcelain inlays were designed and milled from CEREC Blocks (CEREC Blocs PC 14/14 14 S2-PC, item #6038504) using the CEREC inLab MCXL system. The intaglio surfaces of all inlays were treated with 5% hydrofluoric acid etch for 60 seconds, rinsed for 60 seconds, and silinated for 60 seconds. Restorations were cemented to the respective teeth specimens using Nexus NX3 resin cement (Kerr Corp, Orange, CA, USA) per the manufacturer's instructions. Excess cement was removed followed by restoration and margin polishing with diamond-impregnated polishers.

Following restoration placement, all teeth were assessed with a Hirox KH-1300 digital microscope (Hirox, Hackensack, NJ, USA). Specimens were evaluated at 35× magnification to ensure ceramic structural integrity (no ceramic fractures) and at 50× magnification along the gingival margin in a bucco-lingual dimension between the ceramic and tooth structure (enamel and cementum groups) or between the ceramic and margin elevation material (GI or RMGI groups) to verify marginal integrity (completely closed margin with intact resin cement layer without gaps or voids). Additionally, occlusogingival ceramic inlay heights were measured with a digital caliper from the middle of the marginal ridge

in bucco-lingual dimension, down apically to the extent of ceramic at its gingival margin. (Figure 2).

#### Thermomechanical Loading

All teeth underwent thermocycling, to include 10,000 cycles between 5°C and 55°C with 30 seconds of dwell time at each temperature to simulate thermal changes that occur within the oral cavity. Following thermocycling only, structural and marginal integrity were assessed again with the Hirox digital microscope as described above. To simulate mechanical stress on the restorations, all specimens underwent chewing simulation (Chewing Simulator-4, SD Mechatronik, Westerham, Germany). Teeth were mounted in acrylic resin, and the occlusal surface was articulated against an 8-mm stainlesssteel ball antagonist (Figure 3). Fifty newtons of vertical force were applied for 1,200,000 cycles of masticatory simulation. These thermal and mechanical conditions are considered to simulate approximately five years of intraoral service.<sup>35</sup> Following masticatory loading, all samples were again assessed for structural and marginal integrity with the Hirox digital microscope.

#### **Statistical Analyses**

After thermomechanical loading as well as structural and marginal integrity reassessment, statistical analyses were completed. The Fisher exact test (twotailed) was used to compare the four specimen groups, and Bonferroni correction was used to adjust p-values (p<0.05 for statistical significance). Additionally, the association of ceramic height with the probability of ceramic fracture was estimated using logistic regression. Data were analyzed using IBM SPSS Statistics for Windows (version 24.0, IBM Corp, Armonk, NY, USA) and R (version 3.4.2, R Core Team, Vienna, Austria). The null hypothesis was that there was no difference in structural and marginal integrity of ceramic inlays whether cemented to tooth structure or to GI/RMGI deep margin elevation material.

#### **RESULTS**

Following thermocycling alone, ceramic structural and gingival marginal integrity showed no changes. All-ceramic restorations remained intact, and gingival margins remained closed without gaps or visible cement layer discrepancies at 50× magnification (data not shown). After mechanical loading through chewing simulation, the major finding at 35× magnification was the lack of ceramic structural integrity in the cementum margin group. Nine of 10

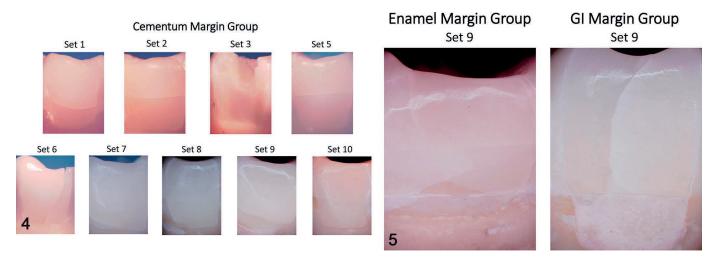


Figure 4. Nine of 10 ceramic inlays in the cementum margin group showed bulk fracture. Varied fracture pattens were present.

Figure 5. Only one of 10 inlays from the enamel margin group and glass ionomer (GI) margin group showed ceramic fracture.

ceramic inlays in the cementum group showed bulk fracture of the ceramic (Figure 4). Only one of 10 inlays from the enamel and GI groups had ceramic bulk fracture (Figure 5), and none of the 10 inlays from the RMGI group had ceramic fracture following thermomechanical loading. Marginal integrity was maintained between ceramic, GI, RMGI, and tooth structure when comparing pre- and postthermomechanical loading images at 50× magnification (Figure 6).

Using the Fisher exact test (two-tailed) and adjusted p-values with a Bonferroni correction to account for multiple comparisons, the ceramic fracture rate for the cementum group was found to be significantly higher than the other three groups (cementum vs enamel: p=0.007; cementum vs GI: p=0.007; and cementum vs RMGI: p<0.001) (Figure 7).

The association of ceramic inlay height with the probability of ceramic fracture was estimated using logistic regression. Ceramic fracture probability increased drastically as occluso-gingival ceramic heights increased. Looking at the actual data and grouping the fracture outcomes into 1-mm increments, no fractures were seen in inlays with heights less than 4.5 mm, 8% fractures in inlays with ceramic heights between 4.5 and 5.5 mm, 29% fractures in teeth between 5.5 and 6.5 mm, and 89% fractures when inlays were greater than 6.5 mm in occluso-gingival height (Figure 8).

#### DISCUSSION

Placing indirect restoration margins on direct restorative materials instead of sound tooth structure is in contradiction to concepts that have been taught for decades, hence the title of Magne's study from 2012: "Deep Margin Elevation: A Paradigm Shift." 19 The DME technique has met resistance due to concerns that failure of margin-elevated restorations arises from the additional restorative material interface between ceramic and direct restorative material. Kielbassa's systematic review on proximal box elevation showed various restorative materials to be successful at maintaining clinically acceptable margins using the DME technique yet still recommended high-quality clinical trials to confirm benchtop outcomes.3 This study also showed that marginal integrity was visibly maintained at 50× magnification across all samples.

Interestingly, one sample group in this study, the cementum margin group, with tall occluso-gingival ceramic inlay heights, demonstrated a lack of ceramic structural integrity that was significantly different than the other three groups. This finding exposes a potential additional benefit of DME beneath ceramic inlays: that the act of placing a direct restoration on the gingival floor inherently shortens the occluso-gingival height of the proximal portion of the inlay. And, based on logistic regression extrapolation of the data found in this study, shorter heights of proximal ceramic inlays are less associated with bulk ceramic fracture and restoration

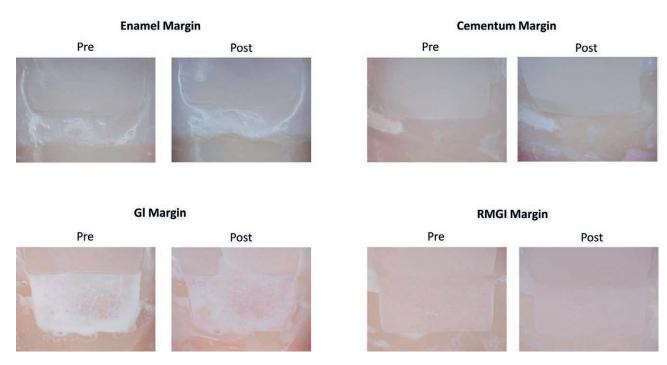
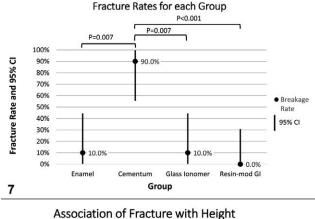


Figure 6. No gingival margin defects before and after thermomechanical loading.



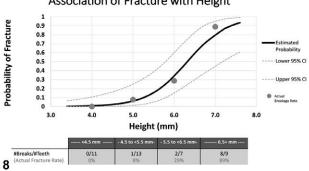


Figure 7. Statistical analysis for fracture rate with 95% confidence interval (CI) between groups.

Figure 8. Logistic regression for the association of ceramic fracture probability with ceramic proximal height.

failure. At five years of simulated function, it appears that occluso-gingival proximal heights of ceramic inlays greater than 5 mm begin to increase bulk fracture rates above the estimated 10% from logistic regression. In other words, to keep survival rates above 90% for ceramic inlays at five years of service, clinicians should consider DME when the proximal box is greater than 5 mm in occluso-gingival height.

Dental anatomy textbooks report the "cervicoocclusal length of crown" of posterior teeth by measuring from a facial view from cusp tip to CEJ. These average heights measure from 8.5 mm in the premolars down to 7.0 mm in the molars.<sup>36</sup> When viewing from the interproximal surface of teeth, these average heights are decreased due to natural anatomic form, going down from cusp tip to marginal ridge as well as the CEJ going up from the midfacial to the interdental position. While no proximal average heights can be estimated, it is likely that "cervico-occlusal length of crown" viewed from the interproximal is encroaching on the potentially critical 5-mm height mentioned in the previous paragraph. Therefore, for clinical ease of visualization, it could be extrapolated that any time an interproximal box preparation for a ceramic inlay ends below the CEJ on dentin, the box will be greater than 5 mm deep and, in turn, potentially could benefit from DME.

The ceramic used in the study, feldspathic porcelain, while highly esthetic due to its high glass content, does not possess fracture resistance similar to natural teeth. Material thickness is required when using these materials to help prevent bulk fracture.<sup>37</sup> Over the years, physical properties of allceramic restorations have improved by adding fillers such as lithium disilicate and alumina to the glass matrix to give greater strength and, in turn, greater fracture resistance.<sup>38</sup> Reported average flexural strengths for some ceramics are as follows: 61 to 87 MPa for feldspathic porcelain, 300 to 500 MPa for lithium disilicate, and 800 to 1200 MPa for yttriastabilized zirconia.8 Although less strong, clinicians continue to use feldspathic ceramic blocks for CAD/ CAM restorations due to esthetics and ease of fabrication, not requiring firing prior to bonding. When comparing milled ceramic vs milled composite inlays, feldspathic ceramic fractures did not involve tooth structure, similar to fracture patterns as were seen in this study, whereas bonded composite restoration fractures more often involved tooth structure.<sup>39</sup> Ceramic restorations seem to concentrate stress within the restoration itself, while composite restorations transfer more stress to tooth structure.35

Also, when comparing direct composite restorations with conventional indirect restorations, Zaruba and others<sup>40</sup> showed that direct composite margins were inferior to those of indirect restorations. Marginal integrity of margin-elevated ceramic indirect restorations from this study was maintained, although further dye staining of margins, sectioning of teeth, and assessing dye penetrance might have revealed additional information regarding marginal integrity maintenance through the simulated five years of service. While the majority of DME studies are in vitro and use resin composite to elevate the margin. 7,20,21,41 the difference in placing a composite in vitro vs in vivo in a difficult-to-access, humid subgingival environment, also complicated by the lack of consensus on the isolation and application technique, should be considered. 41 This study showed that using restorative GI and RMGI for margin elevation gave clinically acceptable marginal integrity at five years of service simulation. With this finding and the aforementioned beneficial properties of GI/RMGI restorative materials, including low modulus of elasticity, coefficient of thermal expansion closest to tooth structure, hydrophilic nature, fluoride release and recharge, and strong chemical bond to tooth structure, practitioners may consider using GI or RMGI instead of resin composite when performing DME. Composite placement is more technique sensitive, has a less predictable bond to dentin, and undergoes polymerization shrinkage and size changes with temperature changes due to its coefficient of thermal expansion, which can lead to microleakage, secondary caries, and restoration failure.

Some clinicians tend to avoid deep DME due to the risk and likelihood of invasion of biologic width when placing restorations that encroach on the crest of bone. Case reports have shown that smooth, nonirritating margins that invade biologic width can be free of gingival and periodontal inflammation provided that meticulous oral hygiene maintenance is performed.<sup>16</sup> Thoughts regarding biologic width have changed over the years since Gargiulo and others<sup>15</sup> proposed the dimensions in 1961. Today, it is believed that there is great variability in biologic width from patient to patient. A recent systematic review looking at this concluded that "no universal dimension of biologic width appears to exist. Establishment of periodontal health is suggested prior to the assessment of biologic width within reconstructive dentistry."42 Overall, pushing the limits of biologic width for DME can potentially save the patient from needing invasive and irreversible removal of bone from surgical crown lengthening.

As is the case with all laboratory-based projects, the results of this laboratory study cannot be directly applied to clinical scenarios. One possible weakness in the study design was that mounted test teeth had no proximal surface from an adjacent tooth to provide support to the ceramic inlays during occlusal loading. Adjacent proximal surfaces from neighboring teeth in the oral cavity may provide some support and distribute chewing forces more evenly. Another drawback is that the chewing simulator provided vertical loading only, whereas in the oral cavity, forces are always multidirectional. Finally, a laboratory study cannot simulate the complexity of the oral environment, nor can it forgo the challenge of isolating the clinical operating field on difficult-to-access posterior tooth preparations. Therefore, randomized controlled clinical trials with appropriate recall intervals are needed to corroborate laboratory findings and substantiate new techniques.

#### **CONCLUSIONS**

This laboratory study demonstrated that DME resulted in decreased ceramic fracture when preparation margins were located below the CEJ. In the

teeth in which the DME technique was used, the cavosurface marginal integrity was maintained between direct and indirect restorations, suggesting that DME is a valid treatment option for clinicians to consider when subgingival margins make indirect restorations a challenge. No difference was found between margin elevation with GI or RMGI.

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#### **Regulatory Statement**

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of the Walter Reed National Military Medical Center. The approval code for this study is 405992.

#### **Disclaimers**

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#### **Conflict of Interest**

The authors of this article certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## The Effect of a Charcoal-based Powder for Enamel Dental Bleaching

MC Franco • JLS Uehara • BM Meroni • GS Zuttion • MS Cenci

#### Clinical Relevance

Charcoal based-powders are not effective for dental bleaching.

#### **SUMMARY**

Charcoal-based dentifrices for dental whitening are a novelty in the market. Manufacturers claim that such charcoal-based products have whitening, remineralization, antimicrobial, and antifungal properties of charcoal in such products. However, there is no substantial scientific evidence for these claims. This laboratory randomized study was designed to evaluate the whitening properties of a charcoal-based toothpowder. A total of 45 bovine dental enamel discs were randomly distributed into three groups (n=15): group 1, mechanical brushing with a 1450-ppm F toothpaste (control group); group 2, mechanical brushing

Marina Christ Franco, PhD student, Restorative Dentistry, Federal University of Pelotas, Rio Grande do Sul, Brazil

Juliana Lays Stolfo Uehara, PhD student, Restorative Dentistry, Federal University of Pelotas, Pelotas, Brazil

Bárbara Meirelles Meroni, graduate student, Federal University of Pelotas, Pelotas, Brazil

Giovanna Sacco Zuttion, graduate student, Federal University of Pelotas, Pelotas, Brazil

\*Maximiliano S Cenci, Graduate Program in Dentistry, Department of Operative Dentistry Federal University of Pelotas, Pelotas, Brazil

\*Corresponding author: R Gonçalves Chaves 457, Pelotas, N/A 96015560, Brazil; e-mail: cencims@gmail.com

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with an activated charcoal-based powder; group 3, bleaching per the standard protocol using 10% carbamide peroxide. The surface roughness and color of each specimen were analyzed at baseline and after 14 days of experiment. The surface of one randomly selected specimen from each group was examined using a scanning electron microscope (SEM). The Kruskal-Wallis test was used to compare groups at a significance level of 5%. Only group 3 promoted a statistically significant effect on  $\Delta E$  compared with groups 1 and 2 (p<0.001 and p=0.003, respectively). No statistically significant difference was found between groups for surface roughness (p>0.05). SEM revealed a more irregular surface in group 1 specimens compared with group 2 and 3 specimens. The charcoal-based powder did not seem to have any bleaching effect.

#### INTRODUCTION

White teeth are regarded as a perfect pattern of esthetic beauty in contemporary society. <sup>1,2</sup> According to data from published literature, 30% of patients have a certain degree of dissatisfaction with their dental color. <sup>3,4</sup> Therefore, the dental bleaching procedure is a popular esthetic treatment in dental practice. <sup>3,4</sup> This procedure is conservative, safe, and effective and is performed using gels containing hydrogen peroxide or carbamide peroxide at differ-

ent concentrations. Dental bleaching can be accomplished at the dental clinic or in the patient's home by following a dentist's instructions. <sup>1,4,5</sup> There are also low-cost alternatives available in the market to treat tooth discoloration without the dentist's supervision. In the last few years, the availability of a number of alternative whitening agents, such as prefabricated trays, whitening strips, and paint-on applications, has considerably increased. <sup>4,6</sup>

The use of charcoal for oral health was recorded for the first time in ancient Greece by Hippocrates. Charcoal was used in many parts of the world and different cultures as a cleaning agent. Natural charcoal powder was used as a single-agent dentifrice. However, studies have shown that charcoal may not be beneficial in dental cleaning or against dental caries.<sup>7,8</sup>

Charcoal-based toothpastes and toothpowders for dental whitening are widely available for purchase from pharmacies, supermarkets, and e-commerce sites. Although manufacturers promise the whitening, remineralization, antimicrobial, and antifungal properties of charcoal-based dentifrices, there is no substantial scientific evidence for these properties. This laboratory study aimed to evaluate whether activated charcoal powder has a bleaching/whitening effect on the dentition. We tested two hypotheses: 1) traditional bleaching procedure with carbamide peroxide is more effective in changing tooth color teeth than whitening with charcoal-based powder and 2) the use of charcoal-based powder increases enamel surface roughness.

#### **METHODS AND MATERIALS**

#### **Experimental Design**

This laboratory randomized study was designed to evaluate the whitening properties of a charcoalbased toothpowder compared with dental bleaching with a gold standard protocol using 10% carbamide peroxide. The control group was treated with a 1450ppm F toothpaste. A total of 45 enamel discs were randomly divided into three groups (n=15): group 1, mechanical brushing using a 1450-ppm F toothpaste (control group); group 2, mechanical brushing using an activated charcoal-based toothpowder; group 3, bleaching protocol with 10% carbamide peroxide. Before- and after-treatment samples were spectrophotometrically evaluated to determine the change in color. In addition, potential deleterious effects of the treatments were assessed by surface roughness changes and scanning electronic microscopy (SEM) images.

#### **Sample Size Calculation**

G\*Power 3.1.9.4 software (Heinrich-Heine Düsseldorf University, Düsseldorf, Germany) was used to determine the sample size based on a previous study<sup>9</sup> using the following parameters: 95% power, 0.40 effect size, and three experimental groups. A minimum sample size of 15 specimens per group (n=45) was assessed to be appropriate.

#### **Specimen Preparation**

Forty-five enamel discs were cut from the buccal surface of fresh bovine incisors using a water-cooled trephine drill. To obtain standardized enamel discs with a diameter of 5 mm and thickness of 2.5 mm, the surfaces were ground using #600-grit silicon carbide papers. Subsequently, all the specimens were polished by wet grinding, sequentially, with #1200-, #1500-, and #2000-grit silicon carbide papers to obtain a polished and standardized surface. The specimens were then numbered and randomly allocated in acrylic matrices, which immobilized them on the brushing machine, according to a list generated by RANDOM.ORG.

#### **Roughness Measurements**

The surface roughness of each specimen was analyzed using a roughnessmeter (Hommel Tester T1000, Hommel-Etamic, Schwenningen, Germany) at baseline and after 14 days of the experiment. Multidirectional readings were made for each specimen from the center of the surface. Three readings were made for each specimen, and the mean was calculated.

#### **Color Stability Evaluation**

The specimen color was assessed at baseline and after 14 days using a spectrophotometer (X-Rite SP60 Series, X-Rite Inc, Grand Rapids, Michigan USA) and D65 light against a white background. To standardize the color measurement, the spectrophotometer pointer was positioned parallel to the enamel surface of each specimen. The shade was determined according to spectrophotometric parameters by considering L\*, a\*, and b\* values. L\* represents values from 0 (black) to 100 (white), a\* represents the amount of red and green, and b\* represents the amount of yellow and blue. The difference in color before and after treatment was provided by delta E ( $\Delta$ E), calculated by CIEDE2000<sup>10</sup>.

Table 1: Components of Products Used According to the Manufacturer's Information			
Product Manufacturer Composition		Composition	
Colgate Maximum anticaries protection	Palmolive Company, New York, NY, USA	1500 ppm of fluoride, calcium carbonate, sodium lauryl sulfate, sodium saccharin, tetrasodium pyrophosphate, sodium silicate, polyethylene glycol, sorbitol, carboxymethyl cellulose, methylparaben, propylparaben, aromatic composition and water; contains sodium monofluorophosphate	
Whitemax	Dermavita, Brusque, SC, Brazil	Charcoal powder (activated carbon), kaolin, aroma, citrus aurantium dulcis peel oil	
Whiteness Perfect	FGM Odontology Products, Joinville, SC, Brazil	Carbamide peroxide, neutralized carbopol, potassium nitrate, sodium fluoride, humectant (glycol), deionized water	

#### **Mechanical Brushing and Whitening Protocol**

Group 1 and group 2 specimens were submitted to mechanical brushing cycles in a Multifunctional Oral Cavity Simulator (Federal University of Pelotas, Pelotas, Brazil). Briefly, this device comprises a Multifunctional Oral Mouth Simulator originally designed to allow continuous flow for biofilm growth with modifications to enable brushing simulation.<sup>11</sup> Brushing simulation was performed with a load of 4.5 N using soft-bristle brushes (Sanifill Eco Dent, Interbros GmbH, Schönau, Germany) at 0.6 Hz (36 cycles/min for three minutes). 12 In group 1 (control group), a toothpaste slurry (Colgate-maximum anticaries protection, 1450 ppm F, Palmolive Company, New York, NY, USA) was prepared with distilled water at a 1:3 (w/v) proportion and applied during the brushing cycles. In group 2, a charcoal-based powder for dental whitening (Whitemax, Barueri, Brazil) was applied on previously wet brushes with water. Group 3 specimens were submitted to a gold standard protocol for dental bleaching with 10% carbamide peroxide (Whiteness Perfect, FGM, Joinville, SC, Brazil) for 3 h/day. The whitening gel was removed after three hours using abundant water. Information of the groups and products used, including manufacturers and product components, is provided in Table 1.

After the bleaching procedure and mechanical brushing cycles, the specimens were stored in distilled water at 37°C for 23 hours until the next

Table 2: Results for 2 Groups <sup>a</sup>	1E Values Based on Tre	eatment
	Median (Quartiles 25%-75%)	Mean (SD)
Control	0.74 A (0.83-1.69)	0.95 (0.51)
Charcoal-based powder	1.24 A (0.83-1.69)	1.28 (0.50)
Carbamide peroxide	2.36 B (2.00-3.60)	2.65 (1.11)
<sup>a</sup> Means followed by equal letters indicate no statistically significant differences in each column (p>0.05).		

exposure to whitening protocol and new brushing. This process was repeated daily for 14 days.

#### **Surface Morphology**

The surface of one randomly selected specimen from each group was examined for finished surface morphology. The specimens were subjected to vacuum in a sputter coater (SCD 050 Sputter Coater, Capovani Brothers Inc, New York, USA) to deposit a thin layer of gold before submitting to SEM (JSM 5600LV, JEOL, Tokyo, Japan).

#### **Statistical Analysis**

The data obtained were double entered and analyzed using SPSS Statistics software (SPSS, Inc, Cary, NC, USA). Descriptive analysis was performed by estimating the mean and SD for all groups. Data of all parameters were examined for normality using the Shapiro-Wilk test. Because the color evaluation data presented nonparametric distributions, the Kruskal-Wallis test was used to compare groups. Surface roughness was evaluated using one-way analysis of variance (ANOVA), followed by Tukey post hoc tests. For all analyses, an  $\alpha$  value of 0.05 was used to determine statistical significance.

#### **RESULTS**

The Kruskal-Wallis test results for  $\Delta E$  are presented in Table 2. Statistical differences were observed between group 3 and group 1 (p<0.001) and between group 3 and group 2 (p=0.003). No statistically significant difference was observed between group 1 and group 2 (p=0.546).

No statistically significant difference was noted between the groups for surface roughness (group 1 vs group 2: p=0.623; group 1 vs group 3: p=0.157; group 2 vs group 3: p=0.613; Table 3).

Figure 1 is an SEM image of enamel surface (A, group 2; B, group 1; and C, group 3). Photomicrographs of representative areas were taken at 35×

Table 3:	Mean, SD, and Confid Roughness (ra) Evalu Groups <sup>a</sup>		
	Mean	SD	Confidence

	Mean	SD	Confidence Interval (95%)
Control	140.14 A	41.37	(117.23-163.05)
Charcoal-based powder	128.57 A	25.36	(114.53-142.62)
Carbamide peroxide	116.80 A	33.33	(98.34-135.26)

 $<sup>^{\</sup>rm a}$  Means followed by equal letters indicate no statistically significant differences in each column (p>0.05).

and 500× magnification for all groups. SEM characterization of the enamel surfaces after different surface treatments revealed a more irregular surface in group 1 specimens (Figure 1B) compared with group 2 and group 3 specimens. Group 2 and group 3 specimens (Figure 1A and 1C, respectively) seemed to have a smooth surface.

#### DISCUSSION

This study showed that the charcoal-based tooth-powder was not effective for dental bleaching, corroborating the first hypothesis. To the best of our knowledge, only one study<sup>13</sup> has assessed the potential whitening effects of charcoal powder to date. Four groups were tested in that study: water (negative control), activated charcoal (experimental group), coconut oil (positive control), and hydrogen peroxide (positive control). The study concluded that activated charcoal and coconut oil were not effective for dental bleaching, corroborating the results of our study.

The at-home dental bleaching under professional supervision using 10% carbamide peroxide and custom trays is safe and presents excellent esthetic results. Therefore, it is considered the gold standard for treating tooth discoloration.<sup>2</sup> Although associated with high cost and need for professional supervision, dental bleaching with 10% carbamide peroxide is the most preferred procedure in dental practice.<sup>14</sup> The interest for low-cost alternatives to dental bleaching and the search for organic and natural ingredients brought attention to charcoal-based preparations.

Charcoal is used for several medical indications, including poisoning and drug overdose. Charcoal is also recognized as a food ingredient and a food coloring agent in oriental countries for its claimed health benefits. <sup>15–17</sup> Its use for dental cleaning has been known for many years in different cultures. <sup>7</sup> In the last few years, a few charcoal-based products were disseminated as an organic and safe alternative to conventional dental bleaching techniques. The use

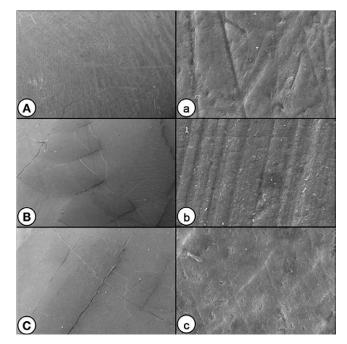


Figure 1. Scanning microscopy of enamel surface per group (A/a group 2; B/b group 1; and C/c group 3) at 35x (uppercase letters) and 500x magnification (lowercase letters).

of these products claimed to result in whiter teeth with affordable cost and without professional supervision. However, there are no studies in the literature on the beneficial effects of charcoal on teeth. $^{7,\ 8}$ 

A few studies have presented inconclusive results regarding the effect of charcoal on oral hygiene, evaluating outcomes for caries, enamel abrasion, halitosis, and periodontal disease. Limited information is available regarding controlled clinical studies that may provide evidence for the use of these products. A laboratory study evaluated the bleaching effect of a charcoal-based toothpaste and demonstrated a small whitening effect. However, this was a subjective color evaluation using the VITA Classical Shade Guide.

In our results, only group 3 presented a statistical difference in the final color for  $\Delta E$  parameters. There was no difference in the final color in group 1 and group 2 specimens. This might be because of the absence of a whitening agent in the charcoal-based powder and the apparent feeling of whiter teeth due to the contrast with the dark color of the powder.

Considering the parameters established by Paravina and others, <sup>18</sup> the acceptability threshold is a  $\Delta E$  of 1.8. Accordingly, only group 3, with a  $\Delta E$  of 2.65, showed a significant color change. According to these

parameters, a  $\Delta E$  of 0.8 is considered clinically perceptible. Thus, the color change promoted by the charcoal-based powder, although not statistically detectable, was clinically perceptible, but it is not comparable to a bleaching effect. This slight color change may have been caused by enamel wear, which typically occurs following the use of abrasive toothpastes and could be easily confused as whitening conferred by the substance. <sup>18–20</sup>

In this study, enamel roughness was also evaluated. We examined whether the abrasiveness of the charcoal-based powder causes alteration in the enamel roughness. However, there was no statistical difference between the groups in enamel roughness. Similar results were found in a review by Demarco and others, which concluded that dental bleaching with 10% carbamide peroxide has no harmful effects on enamel roughness. However, no study to this date has evaluated the effects of charcoal powder on the enamel surface.

SEM revealed a smooth surface in group 2 and group 3 specimens. This might have been due to a greater loss of tooth enamel. Therefore, both treatments seem to promote a certain degree of damage in the tooth enamel. However, SEM is a qualitative analysis, which does not allow data collection. Quantitative surface analysis such as profilometry may present more specific results in such studies.<sup>21</sup>

#### CONCLUSION

The charcoal-based toothpowder had a certain degree of whitening effect, but it was not as effective as dental bleaching. The study results indicate that charcoal might not have any dental bleaching properties. However, further studies are warranted to determine the effect of charcoal on the dental surface.

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#### **Regulatory Statement**

This study was conducted in accordance with all the provisions, guidelines and policies of the Universidade Federal de Pelotas.

#### Conflict of Interest

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature

or kind in any product, service, and/or company that is presented in this article.

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# Fracture Resistance of Remaining Buccal Cusps in Maxillary Premolar Ceramic Onlay Restorations

I Thainimit • P Totiam • K Wayakanon

#### Clinical Relevance

Preparation designs for bondable partial coverage restorations are varied. As little as 1 mm of thickness of a remaining buccal cusp can be kept when restoring maxillary premolars with bondable partial coverage restorations.

#### **SUMMARY**

Indirect partial coverage restorations have become increasingly popular in recent years as new and improved adhesive materials have been developed. These restorations can preserve substantial amounts of tooth structure. However, there are some aspects of indirect partial coverage restorations for which no clear protocol exists. This study investigated the minimal thickness of the nonfunctional cusp that must be left in a bondable ceramic partial coverage restoration in order to resist compressive force. Ninety sound human maxillary premolar teeth were obtained and used in one of the following three ways. Ten of the sound teeth were used as a control without further preparation. Forty other sound teeth had cavities designed and were tested as

Itsalapong Thainimit, DDS, MSD, Restorative Dentistry, Naresuan University, Phitsanulok, Thailand

Potjaman Totiam, DDS, PhD, Restorative Dentistry, Naresuan University, Phitsanulok, Thailand

"unrestored teeth." The remaining 40 sound teeth received not only cavities but also restoration and were tested as "restored teeth." Both the restored group and the unrestored group were prepared either with an overlay or with varying buccal cusp thicknesses of 1, 2, or 3 mm. In total, there were nine experimental groups with 10 in each group (n=10). The prepared teeth were digitally scanned, and the restorations were designed and fabricated from IPS e.max computer-aided design (CAD) software using a CAD/CAM machine (CEREC MC XL, Dentsply Sirona, Bensheim, Germany). The restorations were cemented with resin cement (Panavia V5). All samples underwent thermocycling and dynamic fatigue simulating approximately one year of actual use. All the teeth were then subjected to compressive load until the point of fracture, and the mode of each fracture was analyzed. Results show that the fracture resistance of the restored groups was significantly higher than the nonrestored groups (p<0.001) and the sound teeth (p<0.05). Crucially, this study determined that 1 mm of remaining buccal cusp thickness in bondable partial coverage restorations for maxillary premolars is sufficient to withstand normal use of the tooth without breakage.

<sup>\*</sup>Kornchanok Wayakanon, DDS, MSD, PhD, Restorative Dentistry, Naresuan University, Phitsanulok, Thailand

<sup>\*</sup>Corresponding author: 99 Moo 9 Thapho, Muang, Phitsanulok 65000, Thailand; e-mail: wkornchanok@hotmail.com https://doi.org/10.2341/19-251-L

#### INTRODUCTION

Dental caries as well as fractures in the tooth structure or in dental restorations are common problems found routinely in clinical practice. Restorative treatment needs to be able to repair and reconstruct the integrity, morphology, and functionality of damaged tooth structure. As part of great evolutionary advances in dental materials science, bondable restorative materials have brought many benefits to the process of restorative treatment. Bondable restorative materials are able to strengthen the remaining tooth structure with only small amounts of tooth reduction, leading to increased longevity of restored teeth.<sup>1-3</sup>

Dental adhesive systems play a vital role in current restorative dentistry. Various restorative materials are able to bond to a natural tooth structure that has been prepared with a suitable mechanical or chemical treatment. Adhesive systems and resin-based materials can then be applied. <sup>4,5</sup> These bondable restorations substantially decrease the amount of tooth reduction needed during the cavity preparation process since bondable restorations do not require extensions for retention.

In cases of extensive damage, using direct dental materials for restoration requires more time and more skill than using bondable restorations, so partial coverage restoration is a preferable alternative. The benefits of the partial coverage technique are related to the increased amount of remaining tooth structure. Partial coverage restoration decreases the chance of pulpal and periodontal tissue inflammation and provides more favorable stress distribution to minimize the risk of tooth fracture. Importantly, the majority of the restoration margin is placed on enamel, which serves as an excellent bonding surface for the adhesive system. Longevity of the restoration's retention and functionality is thus increased. 9-12

Table 1: Experimental Groups With 10 Per Group			
Group	Experimental Design	Note	
S	Sound tooth	Control	
N3	Nonrestored buccal thickness 3 mm	Comparative group	
N2	Nonrestored buccal thickness 2 mm	Comparative group	
N1	Nonrestored buccal thickness 1 mm	Comparative group	
N0	Nonrestored occlusal overlay	Comparative group	
R3	Restored buccal thickness 3 mm	Test group	
R2	Restored buccal thickness 2 mm	Test group	
R1	Restored buccal thickness 1 mm	Test group	
R0	Restored occlusal overlay	Test group	

Preparation designs for partial coverage restorations will vary. The designs are related mainly to the particular features of the damaged area. There are no established guidelines for what minimum thickness of remaining cusps in bondable partial coverage restorations is sufficient to withstand normal use of the tooth without breakage. Below this thickness, reduction of the cusp and its inclusion in the restoration is required. The purpose of this study was to evaluate the influence of the remaining cusp thickness on fracture resistance in onlay restorations fabricated with IPS e.max CAD. The null hypothesis was the remaining cusp thickness has no effect on the fracture resistance of the maxillary premolar teeth when restored with ceramic onlays.

#### **METHODS AND MATERIALS**

#### **Sample Preparation**

Ninety sound human maxillary premolar teeth without cracks, restorations, or carious lesions were selected for this study. All the teeth were previously extracted for orthodontic treatment. The teeth were selected for having similar measurements on the occlusal surface: 9.5-10.5 mm bucco-lingual distance, 7.0-7.5 mm mesio-distal distance, and 8.0-8.5 mm occluso-cervical distance. The teeth had been stored in 0.1% thymol solution at room temperature before selection, and all selected teeth were used within three months of extraction. All teeth were embedded in self-cured acrylic resin 3 mm below the cemento-enamel junction (CEJ).

The teeth were then randomly divided into nine groups as shown in Table 1. The nine groups were built around three factors: whether the tooth was sound, whether the tooth was restored, and the thickness of the buccal cusp.

A high-speed hand piece with  $10^{\circ}$  taper diamond burs was used to prepare onlay mesio-occluso-distal cavities with a width and depth of 2 mm and a divergence of  $10^{\circ}$ . The occlusal cavity was continuous with the proximal cavity with an angle of departure of  $120^{\circ}$ . The gingival walls of these proximal cavities were 2 mm below the pulpal floor.

The next step in the cavity preparation was to create the varied thicknesses of the remaining buccal cusp. Those thicknesses were reduced to 3 mm, 2 mm, or 1 mm, or an occlusal overlay following the cuspal incline plane was used. The palatal cusp was reduced by 2 mm following the incline plane to obtain a butt joint margin. All the preparation depths were measured with a periodontal probe.

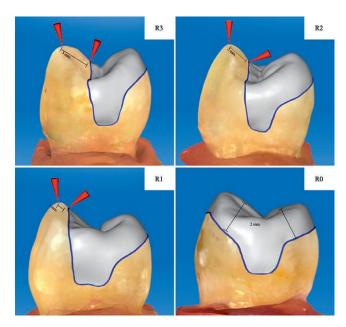


Figure 1. Restoration designs for the four thicknesses of remaining cusp: R3, R2, R1, and R0 (3, 2, 1, and 0 mm, respectively).

#### **Fabrication of the Restorations**

After cavity preparation, the samples were scanned with an intraoral scanner (CEREC Omnicam, Dentsply Sirona, Bensheim, Germany). The restorations were designed with computer-aided design (CAD) software (CEREC SW 4.5, Dentsply Sirona, Bensheim, Germany), re-creating the features of a natural, unprepared tooth (Figure 1). All onlay restorations were milled from lithium disilicate glass-ceramic IPS e.max CAD (Ivoclar Vivadent, Schaan, Liechtenstein) in a milling unit (CEREC MC XL, Dentsply Sirona). After the milling process, heat treatment was performed at a temperature of 840°C for 30 minutes.

#### **Cementing the Restorations**

Before cementation, all the lithium disilicate restorations were examined under a stereomicroscope (SZX-ILLD200, Olympus, Tokyo, Japan) to ensure a lack of any crack or surface flaw. The intaglio surfaces of the restorations were etched with 4.9% hydrofluoric acid (Porcelain Etch, Ultradent, USA) for 20 seconds and then thoroughly rinsed with water for 30 seconds, followed by air-drying. The etched surfaces were then treated with Clearfil Ceramic Primer Plus for five seconds (Kuraray Noritake Dental Inc, Tokyo, Japan) followed again by air-drying.

Phosphoric acid (37%) was applied to the enamel for 15 seconds using the selective etch technique.

After that, the teeth were rinsed with water for 30 seconds, and excess water was removed by airdrying for 15 seconds. Tooth primer was applied and left for 20 seconds, followed by application of selfetch resin cement (Panavia V5, Kuraray Noritake Dental). The self-etch resin cement was handled as recommended by the manufacturer and applied on the intaglio surface of the restorations, which were then seated with finger pressure. The excess cement was removed after tack curing with a light-curing unit (Demi Plus LED, Kerr Corp, Orange, CA, USA) for three seconds on each surface. The restoration cement was then cured for 20 seconds on each surface. Finally, the margins of the restorations were finished and polished with a ceramic polishing kit (Luster for silicate ceramics, Meisinger, Centennial, CO, USA). After cementation, all specimens were stored in water at 37°C for seven days.

#### **Investigation of Fracture Resistance**

All specimens were thermocycled in water (SDC20 HWB332R, Yamatake Honeywell, Tokyo, Japan) between 5°C and 55°C with 15 s dwelling time for 10,000 cycles. This thermocycling represented approximately one year of in vivo functioning. 13 After thermal cycling, a fatigue simulation and fracture resistance test were performed in a universal testing machine (Instron Universal Tester, model 8872; Instron Inc, Canton, MA, USA). For the fatigue simulation, all specimens were subjected to dynamic loading of 127.4 N with a metal sphere of 6 mm in diameter at a frequency of 6 Hz.<sup>14</sup> This fatigue simulation was performed submerged in distilled water at 37°C for 240,000 cycles. In order to evaluate fracture resistance, the specimens were submitted to a shear force on the buccal inclined plane. A metal wedge shape with a blunt rounded end (not a sharp tip) was positioned blunt end down at a 20° angle to the lingual incline plane of the buccal cusp. The wedge thus made contact with both the lingual inclined plane of the buccal cusp and the buccal inclined plane of the lingual cusp (Figure 2). This wedge pressed down on each specimen submerged in distilled water with a crosshead speed of 0.5 mm/min until fracture occurred.

#### **Investigation of the Fracture Mode**

Each fractured specimen was observed under a stereomicroscope (SZX-ILLD200; Olympus) to determine the mode of fracture, which was then identified as one of the two categories described below (modified from Burke and others<sup>15</sup>):

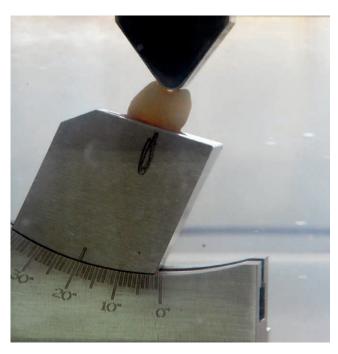


Figure 2. A position of the metal wedge shape was used to shear the specimens at a 20° angle to the lingual incline plane of the buccal cusp.

- Fracture modes by involvement in restoration, dentin, or pulpal tissue
  - Type I: Fracture within the restoration
  - Type II: Fracture of the restoration and tooth structure within the dentin
  - Type III: Fracture of the restoration and tooth structure involving pulpal tissue
- 2. Fracture modes by location above or below the CEJ
  - Type I: Fracture above the CEJ
  - Type II: Fracture below the CEJ

#### **Data Analysis**

The means and standard deviations of the fracture resistance data were calculated and analyzed using two-way analysis of variance (ANOVA) and the Tukey honestly significant difference test in SPSS statistical software (version 23.0, SPSS Inc, Chicago, IL, USA). The significance level was set at 0.05. The fracture mode images were evaluated visually and reported descriptively.

#### **RESULTS**

### Fracture Resistance of Different Cavity Designs

The eight experimental groups (excluding the control) can all be considered permutations of two

	Table 2:	Effect of Preparation Design and Tooth Restoration on Fracture Resistance	
		Factors	<i>p</i> -Value
Ī	Preparation design		0.034
	Tooth restoration		0.000
	Preparation designs_Restorations		0.000

factors: preparation design (buccal cusp thickness 0, 1, 2, or 3) and tooth restoration (restored or not restored). Looking at Table 2, the two-way ANOVA showed that preparation design (p=0.034) and tooth restoration (p<0.001) each had significant effects on the fracture resistance. There was also significant interaction between the preparation design and tooth restoration (p<0.001).

The fracture resistance results are shown in Table 3. Within each cavity design, the restored teeth exhibited significantly higher fracture resistance than the nonrestored teeth, with the exception of the occlusal overlay group (R0 group), in which there was no significant difference in the fracture resistance between the restored and the nonrestored teeth.

Among the restored groups, the R2 group showed the highest fracture resistance (1074.80 $\pm$ 256.91 N). This resistance was significantly higher than the sound teeth (685.67 $\pm$ 187.40 N); however, it was not significantly different from the R1 group (998.69 $\pm$ 279.03 N), the R3 group (986.77 $\pm$ 303.32 N), or the occlusal overlay group (881.64 $\pm$ 234.29 N).

In the nonrestored groups, no significant difference was found among the N1, N2, or N3 groups. The N0 group had significantly higher fracture resistance compared to those other three nonrestored groups (p<0.05), but it was not significantly higher than the sound teeth.

#### **Analysis of the Fracture Mode**

The fracture modes were analyzed by observing the characteristics of the fractured specimens, and the results are shown in Tables 4 and 5 and Figure 3. Looking at whether the various fractures were involved in the restoration, the dentin, or the pulpal tissue (Table 4), the sound teeth and the nonrestored groups exhibited mostly dentin-involved fractures, while the restored groups commonly had pulp tissue—involved fractures. Looking at the same fractures from the perspective of whether they are located above or below the CEJ (Table 5), fractures in the sound teeth and in the nonrestored groups were mostly above the CEJ, except in the N0 group.

Cavity Design	(Group) Fractui	(Group) Fracture Resistance, N		
	Restored	Nonrestored		
Buccal thickness 3 mm	(R3) 986.77 ± 303.32 ABa	(N3) 357.35 $\pm$ 105.36 Ab		
Buccal thickness 2 mm	(R2) 1074.80 ± 256.91 Aa	(N2) 356.05 $\pm$ 177.12 Ab		
Buccal thickness 1 mm	(R1) 998.69 ± 279.03 ABa	(N1) $432.23 \pm 162.19$ ADb		
Occlusal overlay	(R0) 881.64 ± 234.29 ABa	(N0) 983.59 ± 220.22 BEa		
Sound teeth	(S) 685.67 ± 187.40 Ba	(S) 685.67 ± 187.40 CDEa		

In contrast, fractures in the restored group were found for the most part below the CEJ.

#### **DISCUSSION**

There was no statistical difference in fracture resistance when teeth with different remaining cusp thickness (1, 2, and 3 mm) were restored with ceramic onlays. The null hypothesis was not rejected.

The popularity of partial coverage restorations has increased steadily with innovations in adhesive dentistry and in bondable restorative materials. In this study, lithium disilicate ceramics received surface treatment with hydrofluoric acid and silane, <sup>16-18</sup> followed by resin cement application. This process enabled these ceramics to bond to the tooth structure, forming bondable restorations.

The design of this study focused on varying thicknesses of the buccal cusp of the maxillary premolar tooth, in other words, the nonfunctional cusp. According to a previous study, the nonfunctional cusp was more prone to fracture than the functional cusps. <sup>19</sup> Maxillary premolar teeth were intentionally selected for the current study since they have a higher frequency of fracture than do mandibular premolar teeth. <sup>20</sup> Although molar teeth have an even higher fracture rate, they were not

selected for this study because almost all extracted molar teeth are removed due to periodontal problems or dental caries, which would have been undesirable for this study. Extracted molar teeth have also usually been in use for a longer period of time, and they are extracted at a wider variety of ages. Thus, the dentin characteristics of different extracted molar teeth are likely to be dissimilar.<sup>21,22</sup>

Since maxillary premolar teeth are located at the curvature of the jaw and because they have steep cusps, they are naturally exposed to repeated oblique occlusal force. To simulate this natural situation, the loading force in both the fatigue simulation and the fracture resistance tests of this study was applied at a corresponding 20° oblique angle. The resulting fracture resistance of the restored groups in this study was similar to that seen in previous studies, generally around 800-1100 N.<sup>1,24</sup>

Several studies have shown that cusp coverage with a bondable restoration is able to protect a weakened tooth structure. In the current study, the fracture resistance of restored teeth, even those without cusp coverage, was significantly higher than that of unrestored teeth. The bondable restorations, with or without cusp coverage, were able to strengthen the remaining tooth structure. Importantly, as little as a 1 mm thickness of the

Table 4: Fracture Modes by Involvement in Restoration, Dentin, or Pulpal Tissue				
Group	Type I, Within Restoration % (number of samples)	Type II, Within Dentin % (number of samples)	Type III, Involve Pulpal Tissue % (number of samples)	
S	<del>-</del>	100 (10)	0	
N3	<del></del>	100 (0)	0	
N2	<del></del>	100 (0)	0	
N1	<del></del>	100 (0)	0	
N0	<del>-</del>	90 (9)	10 (1)	
R3	0	0	100 (10)	
R2	10 (1)	20 (2)	70 (7)	
R1	20 (2)	30 (3)	50 (5)	
R0	0	20 (2)	80 (8)	

Table 5:	Fracture Modes by Location Above or Below the Cemento-Enamel Junction (CEJ)	
Group	Type I, Above CEJ % (number of samples)	Type II, Below CEJ % (number of samples)
S	100 (10)	0
N3	100 (10)	0
N2	90 (9)	10 (1)
N1	80 (8)	20 (2)
N0	40 (4)	60 (6)
R3	0	100 (10)
R2	10 (1)	90 (9)
R1	50 (5)	50 (5)
R0	0	100 (10)

remaining nonfunctional cusp of a maxillary premolar can be kept without reduction.

This study showed that differences in preparation design or buccal wall thickness had considerable influence on the fracture resistance of the maxillary premolars. The fracture resistance of the reducedcusp restorations (overlays) was not significantly different from that of the nonreduced ones. Guess and others similarly found that preparation design and ceramic thickness influenced the fracture resistance of premolar partial coverage restorations. 1 However, extending preparation from a palatal onlay to complete occlusal coverage on premolars did not increase the fracture resistance. A clinical study conducted by Van Dijken and others<sup>27</sup> found no statistically significant difference in performance between four preparation designs, even five years after ceramic partial coverage restorations. Similar results have been observed in other in vitro studies.<sup>24,28,29</sup>

After cementation, this study used thermal cycling and dynamic fatigue to simulate actual clinical conditions because humidity in the oral environment, along with the mechanical and thermal stress, has been shown to affect fracture resistance of restorations.<sup>30</sup> The mean fracture resistance of nonrestored teeth determined in the current study was lower than that found in some other studies.<sup>31,32</sup> The specimens in this study received dynamic fatigue and loading force underwater, while the previous studies performed the fatigue under dry conditions. Many other variables can also affect fracture resistance, such as the mode and direction of loading as well as the crosshead speed and size of the indenter.<sup>15,29</sup>

All of the ceramic partial coverage restorations in this study had a greater fracture resistance than the



Figure 3. These photographs show three fractured teeth (horizontally) from three perspectives (vertically). The fractures can be classified by whether they are involved in the restoration (A, D, and G), dentin (B, E, and H), or pulpal tissue (C, F, and I). They can also be classified by location above (A, D, and G) or below (B, C, E, F, H, and I) the cemento-enamel junction.

physiological masticatory forces to which they would typically be exposed. The average occlusal forces in the first premolar area has previously been measured as  $178.54\pm77.20~N~(for~women)$  and  $254.08\pm72.20~N~(for~men).^{33}$  In a 2011 study, the occlusal forces of premolars were measured as  $373.8\pm102.6~N~for~men$  and  $314.7\pm96.5~N~for~women.^{34}$  Thus 400~N~is~a reasonable estimate of the minimum threshold of occlusal force to which a premolar tooth is normally subjected.  $^{31}$ 

This study assessed fracture mode from two perspectives: whether the fracture is involved in the restoration, the dentin, or the pulpal tissue or whether the fracture is located above or below the CEJ. Among the restored groups, the majority of fracture modes involved pulpal tissue and extended below the CEJ into the radicular portion. On the other hand, among the nonrestored groups, most fractures were within the dentin and above the CEJ. The restored groups had fracture resistance three times higher than the nonrestored groups. This could be explained by additional strength provided by the bondable ceramic restorations.

Resin cement has the dual benefits of esthetic appeal and bond strength, whether applied to the dental structure or to the ceramic restoration. Resin cement also enhances the flexural strength of a tooth restored with bondable stiff material, such as

ceramic.<sup>24,35</sup> Because the force produced by actual chewing is less than the force that was applied experimentally in the laboratory, these restorations should be able to withstand normal mastication.

Some limitations of this study were associated with the mechanical testing of the extracted teeth. The single direction of force applied to the teeth is not a load typically occurring in actual mastication. The mechanical test was also not able to account for absent periodontal structure, which would provide some natural shock absorption.

#### **CONCLUSIONS**

This study succeeded in evaluating the influence of the remaining cusp thickness on fracture resistance in restorations fabricated with IPS e.max CAD onlays. Crucially, it determined that a 1 mm thickness of a remaining buccal cusp in bondable partial coverage restorations is sufficient to withstand normal use of the tooth without breakage. Other sections of this study were able to clarify outstanding questions about designing partially covered restorations conservatively, information that should be of good use during treatment.

#### Acknowledgements

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#### **Regulatory Statement**

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of the Naresuan University. The approval code issued for this study is 0501/61.

#### **Conflict of Interest**

The authors of this article certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Influence of Tooth Pigmentation on H<sub>2</sub>O<sub>2</sub> Diffusion and Its Cytotoxicity After In-office Tooth Bleaching

CC de Oliveira Duque • DG Soares • ALF Briso • U Ortecho-Zuta RA de Oliveira Ribeiro • J Hebling • CA de Souza Costa

#### Clinical Relevance

Pigments in tooth structures affect the diffusion of  $H_2O_2$  through enamel and dentin. The bleaching methodology can be impacted.

#### **SUMMARY**

Objective: The aim of this study was to evaluate the influence of the presence of pigments in tooth structures on the trans-enamel and trans-dentin diffusion of hydrogen peroxide  $(H_2O_2)$  and its cytotoxicity after carrying out an in-office bleaching therapy.

Methods and Materials: A bleaching gel with 35%  $H_2O_2$  was applied for 45 minutes (three times for 15 minutes) on enamel and dentin

Carla Caroline de Oliveira Duque, PhD student, Department of Dental Materials and Prosthodontics, Araraquara School of Dentistry, University Estadual Paulista-UNESP, Araraquara, SP, Brazil

Diana Gabriela Soares, professor, Department of Operative Dentistry, Endodontics and Dental Materials, Bauru School of Dentistry, University de São Paulo-USP, Bauru, SP, Brazil

André Fraga Briso, professor, Department of Restorative Dentistry, Araçatuba School of Dentistry, University Estadual Paulista–UNESP, Araçatuba, SP, Brazil

Uxua Ortecho Zuta, PhD student, Department of Dental Materials and Prosthodontics, Araraquara School of Dentistry, University Estadual Paulista-UNESP, Araraquara, SP, Brazil

Rafael Antonio de Oliveira Ribeiro, MS student, Department of Dental Materials and Prosthodontics, Araraquara School of Dentistry, University Estadual Paulista—UNESP, Araradiscs (n=6), either previously submitted to the intrinsic pigmentation protocol with a concentrated solution of black tea, or not, defining the following groups: G1, unbleached untreated discs (control 1); G2, unbleached pigmented discs (control 2); G3, bleached untreated discs; G4, bleached pigmented discs. The discs were adapted to artificial pulp chambers, which were placed in wells of 24-well plates containing 1 mL culture medium (Dulbecco's modified Eagle's medium [DMEM]). After applying the bleaching gel on enamel, the extracts (DMEM + components of bleaching gel that diffused through the discs) were collected and then applied on the cultured MDPC-23 odontoblast-

quara, SP, Brazil

- Josimeri Hebling, professor, Department of Orthodontics and Pediatric Dentistry, Araraquara School of Dentistry, University Estadual Paulista-UNESP, Araraquara, SP, Brazil
- \*Carlos Alberto de Souza Costa, professor, Department of Physiology and Pathology, Araraquara School of Dentistry, University Estadual Paulista-UNESP, Araraquara, SP, Brazil
- \*Corresponding author: Rua Humaitá, 1680, Centro, Araraquara, SP 14801-903, Brazil; e-mail: casouzac@foar. unesp.br

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like cells. Cell viability (methyl tetrazolium assay and Live & Dead, Calcein AM, and ethidium homodimer-1 [EthD-1] probes), the amount of  $\rm H_2O_2$  that diffused through enamel and dentin (leuco-crystal violet product), and the  $\rm H_2O_2$ -mediated oxidative cell stress (SOx) and components of degradation were assessed (analysis of variance/Tukey;  $\alpha$ =0.05).

Results: There was no significant difference between the groups G1 and G2 for all the parameters tested (p>0.05). Reduction in the trans-enamel and trans-dentin diffusion of  $\rm H_2O_2$  occurred for G4 in comparison with G3. Significantly lower cell viability associated with greater oxidative stress was observed for G3 (p<0.05). Therefore, in-office tooth bleaching therapy performed in pigmented samples caused lower cytotoxic effects compared with untreated samples submitted to the same esthetic procedure (p<0.05).

Conclusion: According to the methodology used in this investigation, the authors concluded that the presence of pigments in hard tooth structures decreases the trans-enamel and trans-dentin diffusion of  $H_2O_2$  and the toxicity to pulp cells of an in-office bleaching gel with 35%  $H_2O_2$ .

#### INTRODUCTION

Tooth bleaching is a clinical procedure with great impact on esthetic dentistry and is the first alternative recommended for the treatment of chromatic changes originating in dental tissue, changes which result in alteration of light reflection and compromised beauty of the smile.

Briso and others<sup>1</sup> reported that the ease of performing the technique, preservation of enamel structure, and obtaining relatively fast clinical results are some of the benefits that have made the use of the in-office bleaching technique the procedure that is still preferred by many patients and professionals. However, the option to use highly concentrated bleaching gels has generated innumerable controversies, and reports of pulp tissue compromise in vital teeth submitted to treatments based on the use of in-office gels containing high concentrations of peroxide have been reported. 2,3 In this context, hypersensitivity is the main adverse effect. 1,4-6 Recent clinical studies have demonstrated that postbleaching effects may be associated with the trans-dentin diffusion of the hydrogen peroxide molecule  $(H_2O_2)$ , the main active component of bleaching gels, and the byproducts of its degradation. These effects may range from inflammation of different intensities to the formation of areas of necrosis in the pulp.<sup>2,3,7</sup>

Bleaching of the tooth structure is known to occur due to the high oxidant power of H<sub>2</sub>O<sub>2</sub> and its reaction products, which have the potential to break polypetide chains and organic components, including the chromophore agents present in the hard tissues of the tooth.8-11 However, when bleaching gels containing high concentrations of H<sub>2</sub>O<sub>2</sub> are used, unreacted (with chromophores) molecules can diffuse through the dentin tubules into the pulp chamber. Unreacted residual H<sub>2</sub>O<sub>2</sub>, being a toxic reactive species, may decrease pulp cell viability and cause oxidative stress and damage to the membranes of these cells.<sup>2,3,12-15</sup> Recent studies demonstrated that the intensity of the toxic effects of H2O2 is inversely proportional to the thickness of tooth enamel and dentin. 11 It is also directly related to the peroxide concentration and time of application used in professional in-office bleaching procedures. This means that the higher the concentration of H<sub>2</sub>O<sub>2</sub> in the bleaching gel and the longer the time of contact of the product with enamel, the more intense will be the adverse cytotoxic effects. 13,15 Based on the mechanism of action, the hypothesis arose that, in dental tissues with a high level of pigment saturation, the pigments might react with a larger quantity of H<sub>2</sub>O<sub>2</sub> coming from bleaching products, thereby reducing the trans-enamel and trans-dentin diffusion of this toxic molecule. Confirmation of this hypothesis seems to be relevant and would allow individualized and safe bleaching therapies to be established, preventing pulp damage and the occurrence of painful postbleaching symptoms. 6,16

Thus, the stated hypothesis of this study was that elevated dentin pigment decreases  $H_2O_2$  diffusion into the pulp to reduce cell cytotoxicity. For this purpose, the present *in vitro* study assessed the influence of intrinsic pigmentation induced in dentin on the trans-enamel and trans-dentin diffusion of  $H_2O_2$ , as well as the toxic effects of this reactive oxygen species (ROS) to odontoblast-like MDPC-23 pulp cells.

#### **METHODS AND MATERIALS**

#### **Experimental Design**

This investigation presented the following factors associate with the study: dental pigmentation at two levels (natural pigmentation and natural pigmentation + tea) and surface treatment at two levels

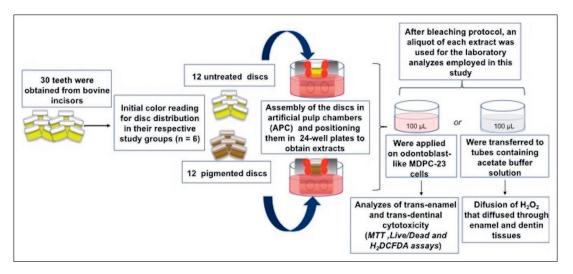


Figure 1. Diagram of experimental design of this research.

(bleaching gel with 35%  $H_2O_2$  and unbleached substrate).

The response variables were trans-enamel and trans-dentin diffusion of  $H_2O_2$  and the cytotoxicity of the treatment evaluated by cell viability tests (methyl tetrazolium assay, MTT), oxidative stress (carboxy- $H_2DCFDA$  probe), and lesion of the cellular membrane (Calcein AM and ethidium homodimer-1 [EthD-1] probes). Twenty-four dental discs were used to perform the protocols (n=6 for each study group) as shown in Figure 1.

#### **Preparation of Samples**

Preparation of Enamel and Dentin Discs—A total of 30 enamel and dentin discs were obtained from the vestibular surface of bovine incisors. The teeth were cut with a diamond-coated trephine bur (Dinser Brocas Diamantadas LTDA, São Paulo, SP, Brazil), with an internal diameter of 5.6 mm, coupled to a bench drill (FSB 16 Pratika, Schultz, Joinville, SC, Brazil), in an environment submersed in water at 4°C. To standardize the thickness of the discs at 3.5 mm, the dentin surface was abraded with 400- and 600-grit water abrasive papers (T469-SF-Norton, Saint-Gobam Abrasivos Ltda, Jundiaí, SP, Brazil). The enamel and dentin sample thickness average was  $0.91 \pm 0.21$  and  $2.59 \pm 0.13$  mm, respectively. Measurement of the amount of enamel and dentin was performed using photographs taken by a DSLR camera (Canon T6i, F 22, ISO 200, speed 180, Otã, Tokyo, Japan). The images were analyzed using Image J software (National Institute of Health, Bethesda, MD, USA). After this, all the enamel surfaces of the discs were submitted to prophylaxis with pumice stone and water. Subsequently, the

dentin surface was treated with ethylenediamine tetra-acetic acid (EDTA 0.5 N, Sigma-Aldrich Corp, St Louis, MO, USA) for 30 seconds to remove the smear layer. Then, the discs were submitted to a colorimetric analysis to perform the first selection of 24 standardized samples, such as described in detail below

First Selection of Discs—Before the experimental protocol, the first selection of discs was made based on initial color. For this purpose, the discs were positioned in an ultraviolet-visible (UV-Vis) reflectance spectrophotometer (Color Guide 45/0, BYK-Gardner GmbH, Geretsried, Germany) with a wavelength ranging from 400 to 700 nm. The color model of the CIE L\*a\*b\* system was used as established by the Commission Internationale de l'Eclairage (CIE; International Commission on Illumination), which allows specification of the perception of colors of the tridimensional model. Three readouts were taken on the vestibular surface of discs. 12,15 After obtaining the L\* and b\* values, the mean value of the entire sample was calculated, and at this time, 24 discs that had L\* and b\* values closest to the mean values were selected. The mean of the L\* values among the selected specimens was 60.87 (range, 58.46-61.71), whereas the mean of b\* was 5.09 (range, 0.83-8.55). It is worth emphasizing that the axes L\* and b\* were considered because the chromatic changes generated by the bleaching treatment occurred preponderantly in these axes.

Pigmentation and Second Selection of Discs—At this time, half of the samples were submitted to an artificial pigmentation protocol, <sup>17</sup> and the other half of the samples remained with their natural color. Thus, as previously described by Moreira and

Table 1: Experimental Groups According to Bleaching Get Concentration (n=6)			
Group	Sample	Treatment	
G1	Untreated unbleached discs	Without treatment	
G2	Pigmented unbleached discs	Without treatment	
G3	Untreated bleached discs	Gel with 35% H <sub>2</sub> O <sub>2</sub> applied three times for 15 min	
G4	Pigmented bleached discs	Gel with 35% H <sub>2</sub> O <sub>2</sub> applied three times for 15 min	

others, <sup>19</sup> 12 discs were incubated in a black tea infusion (0.16 g/mL) at 37°C for six days, with the solution changed every 48 hours. After the staining period, prophylaxis with pumice stone was performed on the enamel surface, and afterward, the samples were incubated in deionized water for 24 hours to eliminate the pigments not absorbed by dentin. The pigmented discs were again positioned in the UV-Vis reflectance spectrophotometer, and a chromatic change of  $\Delta E = 8.77 \pm 1.25$  was found. Also obtained was the mean of L\* (53.50; range, 51.81 to 59.33) and b\* (9.00; range, 1.53 to 11.97).

#### **Cell Culture**

Immortalized MDPC-23 cells were obtained from the stock of cells of the Laboratory of Experimental Pathology and Biomaterials, Araraquara School of Dentistry, UNESP, SP, Brazil. These cells were seeded in 96-well plates (Corning Inc, New York, NY, USA) and incubated in Dulbecco's modified Eagle's culture medium (DMEM; GIBCO, Grand Island, NY, USA), containing 10% fetal bovine serum (FBS; GIBCO), 100 IU/mL penicillin, 100 µg/mL streptomycin, and 2 mmol/L glutamine (GIBCO) in an atmosphere humidified at 37°C, with 5% CO<sub>2</sub> and 95% air, to obtain a standard of 80% of confluence. 12

#### **Experimental Procedure**

The enamel/dentin discs were placed in artificial pulp chambers (APCs), kept in place between two silicone rings, and peripherally sealed with utility wax (Polidental, Cotia, SP, Brazil). Then, the disc/APC sets were sterilized in ethylene oxide (Acecil, Central de Esterilização Comércio e Indústria Ltda, Campinas, SP, Brazil). Afterward the sets were individually distributed in 24-well plates (Corning Inc) containing 1 mL DMEM without FBS. The culture medium remained in contact with the dentin, wheras the enamel was kept exposed either to receive bleaching gel or not, as demonstrated in Table 1. The discs were distributed into the following groups (n=6): G1, unpigmented and unbleached

discs; G2, pigmented and unbleached discs; G3, unpigmented and bleached discs; G4, pigmented and bleached discs. For the bleaching procedure, the product Whiteness HP 35% (FGM Produtos Odontológicos, Joinville, SC, Brazil) was manipulated by adding three drops of the liquid phase containing H<sub>2</sub>O<sub>2</sub> to one drop of thickener (3:1). After this, 20 µL of the resulting gel was applied to the enamel for 15 minutes, and the residual gel was aspirated using a sterilized cannula connected to a vacuum pump. This procedure was repeated two more times to yield a total of 45 minutes of exposure; this is consistent with bleaching protocols used in prior studies. 6,12-15,21 At the conclusion of the bleaching procedure, the extract (DMEM + bleaching gel components that diffused through the enamel and dentin) was collected, homogenized, and distributed into aliquots of 100 µL, which were applied on MDPC-23 cells seeded in 96-well plates (Corning Inc). 12 The cells were incubated for one hour in contact with the extracts to perform the cytotoxicity analyses, by means of the cellular viability tests (MTT), oxidative stress (carboxy-H<sub>2</sub>DCFDA probe), and cellular membrane lesion (Calcein AM and EthD-1 probes). For quantification of the transenamel and trans-dentin diffusion of the H<sub>2</sub>O<sub>2</sub> molecule, an aliquot of 100 µL of the extracts was transferred to a 24-well plate containing acetate buffer solution for stabilization of the H<sub>2</sub>O<sub>2</sub> molecule.

Cell Viability (MTT Assay; n=6)—After having been exposed to the extracts for one hour, the cells were incubated at 37°C and 5% CO2 for four hours in 90 µL DMEM culture medium, to which 10 µL of the MTT solution (3-4,5-dimethylthiazol-2-yl-2,5-diphenyltetrazolium bromide, Sigma-Aldrich Corp) was added, as previously described. 12 After this period, the formazan crystals resulting from the reduction of the tetrazolium salt by the succinate dehydrogenase present in the mitochondria of viable cells were dissolved in 100 µL isopropanol solution acidified in HCl 0.04 N, and the absorbance was measured at 570 nm (Synergy H1, Biotek, Winooski, VT, USA). The mean absorbance value obtained in group G1 was considered 100% cellular viability, and the percentage viability of the other experimental groups was calculated from this parameter. 12-15

Oxidative Stress (Carboxy- $H_2DCFDA$  Probe; n=6)—As previously described,  $^{12,15}$  the cells seeded in 96-well plates (Corning Inc) were pretreated with the fluorescent probe carboxy- $H_2DCFDA$  (Invitrogen, San Francisco, CA, USA) in a concentration of 5  $\mu M$  at 37°C for 30 minutes and were then exposed to the extracts. Immediately afterward, fluorescence

Table 2:	Power of the Statistical Analysis of Cell Viability
	(MTT Assay)

	р	Observed Power
Enamel pigmentation	< 0.001	1.00
Enamel bleaching	0.008	0.888
Interaction	0.100	0.375

was evaluated at 492-nm wavelength excitation and 517-nm wavelength emission (Synergy H1, Biotek), and the values were normalized by the mean values of group G1.

Cell Membrane Disruption (Calcein AM and EthD-1 Probes; n=6)—For this qualitative analysis, the Live/Dead kit (Invitrogen) was used, <sup>12</sup> which is based on the use of two fluorescent markers. After exposure to the extracts, the cells seeded in 96-well plates (Corning Inc) were washed in PBS and incubated with culture medium supplemented with Calcein AM and EthD-1, in a concentration of 1:1000 for 45 minutes. Subsequently, the samples were evaluated under a fluorescence microscope (Leica DM 5500B, Nussloch GmbH, Nussloch, Germany) to obtain representative images of each group.

### Quantification of Hydrogen Peroxide Diffusion $(H_2O_2 \mu g/mL)$

The quantity of H<sub>2</sub>O<sub>2</sub> that diffused through the enamel and dentin was determined by the reaction of H<sub>2</sub>O<sub>2</sub> with leuco-crystal violet dye (0.5 mg/mL, Sigma Chemical Co, St Louis, MO, USA) and with the horseradish peroxidase enzyme (1 mg/mL, Sigma Chemical Co), according to the methodology described by Mottola and others.<sup>22</sup> As previously mentioned,  $^{12,15}$  an aliquot of 100  $\mu L$  of the extract of each experimental group (n=6) was transferred to a plate with 900 µL acetate buffer solution for stabilizing the H<sub>2</sub>O<sub>2</sub>. Afterward, 500 μL of this solution (buffer solution plus extract) was transferred to test tubes containing water and leuco-crystal violet dye (0.5 mg/ mL, Sigma Chemical Co). The tubes were shacked, and 50 µL of the solution at 1 mg/mL horseradish peroxidase enzyme (Sigma Chemical Co) was added. The final volume of the reaction was adjusted to 3 mL with distilled water. An aliquot of 100 µL of each

Table 3: Power of the Statistical Analysis of Oxidative Stress (H<sub>2</sub>DCFDA Probe)

,		
	р	Observed Power
Enamel pigmentation	< 0.0001	1.00
Enamel bleaching	0.008	0.807
Interaction	0.020	0.674

Table 4: Power of the Statistical Analysis of Quantity of  $H_2O_2$  ( $\mu g/mL$ )

	р	Observed Power
Enamel pigmentation	< 0.0001	1.00

solution was transferred to wells of 96-well plates (Corning Inc). The absorbance of the solutions was measured in a spectrophotometer at a wavelength of 596 nm (Synergy H1, Biotek). The standard curve of known quantities of  $\rm H_2O_2$  was used for converting the optic density values of the samples obtained in micrograms of  $\rm H_2O_2$  per milliliter of extract.

#### **Statistical Analysis**

Sample size was based on previous publications  $^{12-15}$  (n=6). The cellular viability and oxidative stress data were normally distributed (Shapiro-Wilk, p>0.05); however, only cell viability data were homoscedastic (Levene, p=0.304). Therefore, these data were submitted to a two-way analysis of variance (ANOVA) considering the factors enamel bleaching and enamel pigmentation, complemented by pairwise comparisons with Tukey's test. The Games-Howell post hoc test was applied to oxidative stress data. Student's ttest was used for analyzing the quantity of  $H_2O_2$  in the extracts of groups G3 and G4. Statistical inferences were taken at the 5% level of significance. The power of the statistical test is shown in Tables 2-4.

#### **RESULTS**

#### Trans-enamel and Trans-dentin Cytotoxicity

For groups G3 and G4, postbleaching cellular viability was reduced by approximately 69.2% and 50.5%, respectively, in comparison with their control groups, G1 and G2 (p < 0.05). In addition, the presence of black tea pigments in dentin (G2) was found to cause no cytotoxicity to the MDPC-23 odontoblast cells (Figure 2). The cells belonging to groups G3 and G4 underwent significant oxidative stress compared to those of G1 and G2 (p < 0.05); Figure 3). However, for the pigmented and bleached discs (G3), there was a reduction of approximately 24% in cellular oxidative stress in comparison to the unpigmented and unbleached discs (G4) (p<0.05). These data were correlated by means of analysis of the images obtained from the live/dead test, in which a large quantity of cells were marked positively with EthD-1 in G3 and G4 in comparison to G1 and G2. A larger quantity of cells marked positively with EthD-1 was detected in G3 compared to G4 (Figure 4).

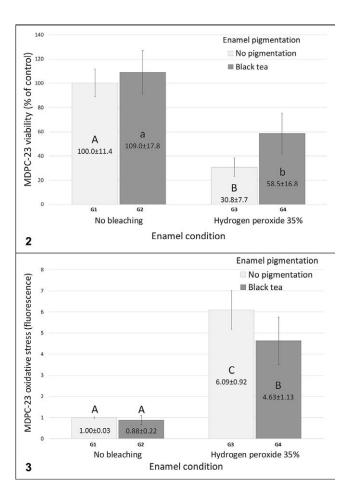


Figure 2. MDPC-23 viability after contact with extracts produced by the application of 35% hydrogen peroxide gel on unaltered or black tea pigmented enamel. Unbleached enamel was used as control. Columns represent means, and error bars indicate standard-deviations (n=6). There was no interaction between the factors (p=0.100), but individually each factor had a significant effect on cell viability (enamel condition and enamel pigmentation, p<0.001). Therefore, columns connected by brackets within the same enamel condition were statistically different. Within the same enamel pigmentation without pigmentation (G1 versus G3) and different lowercase letters allow comparison between the groups without pigmentation (G1 versus G3) and different lowercase letters allow comparison between the groups pigmented with black tea (G2 versus G4). Distinct letters indicate statistical difference between the groups.

Figure 3. Oxidative stress detected in MDPC-23 after contact with extracts produced by the application of 35% hydrogen peroxidase gel on unaltered or pigmented enamel. Unbleached enamel was used as control. Columns represent means, and error bars indicate standard deviations (n=6). There was interaction between the factors (p=0.020); therefore, columns identified with distinct letters are statistically different.

### Quantification of Hydrogen Peroxide Diffusion ( $H_2O_2 \mu g/mL$ )

A high level of trans-enamel and trans-dentin diffusion of  $H_2O_2$  occurred in both groups in which the discs were bleached with gel containing 35%  $H_2O_2$  (G3 and G4). However, this diffusion of  $H_2O_2$ 

through the hard-dental tissues was significantly lower for the discs that were pigmented and then bleached (G4) in comparison with the untreated bleached discs (G3) (p<0.05). The volume of  $\rm H_2O_2$  that reached the pulp space was 6.68 and 4.74 µg/mL for G3 and G4, respectively (Figure 5).

#### DISCUSSION

The chromophores that may accumulate in tooth hard tissues are organic structures formed by aromatic compounds with the presence of amino acids.<sup>23</sup> Among them, tyrosine, tryptophan, and phenylalanine are more expressive with regard to the excitation of fluorophores present in the tooth.<sup>23</sup> In this research, we sought to assess the influence of black tea-induced tooth pigmentation on the transenamel and trans-dentin diffusion and cytotoxicity of the components of a commercial bleaching gel with a high concentration of H<sub>2</sub>O<sub>2</sub> (35%). For this purpose, enamel and dentin discs were submitted to a protocol of intrinsic pigmentation with black tea before the bleaching procedure. Previous studies demonstrated that performing a professional bleaching protocol (35% H<sub>2</sub>O<sub>2</sub> gel, three times for 15 minutes) onto enamel/dentin discs with 3.5-mm thickness, simulating maxillary central incisors, dramatically reduced the dental human pulp cell viability and caused intense changes in cell morphology. 12,13,15,19 However, for cytotoxicity analyses, these in vitro methodologies did not use pigmented samples. To understand the influence of the presence of staining agents on H<sub>2</sub>O<sub>2</sub>-mediated trans-enamel and transdentin cytotoxicity, the enamel/dentin discs were stained with black tea before carrying out the bleaching procedure. This dye was chosen because it is widely used in the literature to evaluate in-office bleaching therapies. 12,13,15,19 In addition, organic pigments present polyphenolic compounds in their structural formulation that are responsible for color sedimentation when in contact with organic substrate. 11,24-28 Despite the limitations of the present investigation, in which only black tea was used for enamel/dentin pigmentation, one should be aware that this pigment is quite representative. This is because it has a similar constituent chemical nature characterized by the presence of polyphenolic compounds, which differ among each other according to the number of phenol units present in the molecule structure.<sup>28</sup>

The test used to evaluate the trans-enamel and trans-dentin diffusion of  $H_2O_2$  demonstrated that, for the pigmented and unbleached teeth (G4), a reduction of approximately 29% in the quantity of

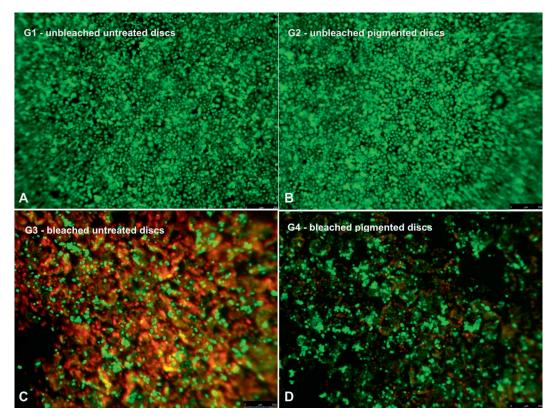


Figure 4. Representative images of live/dead test for each group (A-D). When the cell membrane is ruptured, the EthD-1 marker penetrates into the cell and binds to the nucleotides, emitting fluorescence (red). This marker is incapable of crossing the whole cell membrane. The Calcein AM marker is capable of emitting fluorescence in cells with cytoplasmic esterase activity, irrespective of the cellular membrane condition (green).

these diffused molecules occurred compared with values in the untreated bleached teeth (G3). These data corroborated the findings of the research of Moreira and others, <sup>19</sup> in which the authors observed

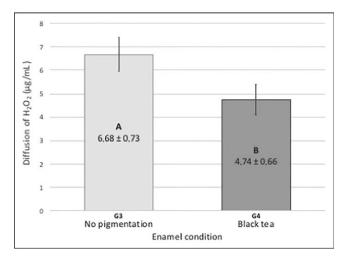


Figure 5. Analysis of residual trans-enamel and trans-dentin  $H_2O_2$ : diffusion. Bar graph of mean values (numbers) and error bars indicate standard deviations of  $H_2O_2$  ( $\mu g/mL$ ) present on the extracts (n=6). Different letters show statistically significant difference among groups (Student's t-test; p<0.0001).

that the presence of H<sub>2</sub>O<sub>2</sub> that did not react with the pigments (residual H2O2) in dentin was approximately 6.25 µg/mL for untreated bleached discs and approximately 5.23 µg/mL for the pigmented bleached discs. In the study of Moreira and others, <sup>19</sup> the authors used bleaching gel with 38% H<sub>2</sub>O<sub>2</sub> and observed trans-enamel and trans-dentin diffusion of this toxic molecule into dentin substrates that presented different intensities of staining. However, after the third bleaching session, the H<sub>2</sub>O<sub>2</sub> diffusion values were statistically similar for all the groups. These data agree with the results obtained in the present research, because it was possible to demonstrate that the presence of pigmentation in the dental substrate significantly minimized the diffusion of residual H<sub>2</sub>O<sub>2</sub>, thus considerably reducing the cytotoxicity caused by the in-office bleaching procedure tested. Therefore, the stated hypothesis of this study was accepted.

The decrease in cellular cytotoxicity is directly related to the reduction in the rate of penetration of residual  $H_2O_2$  diffused through enamel and dentin. <sup>19</sup> Therefore, we believe that the presence of chromophores of pigmentation in tooth substrates and their

interaction with H<sub>2</sub>O<sub>2</sub> could limit the quantity of this molecule that would reach the pulp cells. However, the toxic effect of H<sub>2</sub>O<sub>2</sub> may be of higher or lower intensity, varying according to the organic structure of the chromophore on which it is applied. In this research, the pigmentation agent used was black tea, which has antioxidant properties that make it highly reactive to peroxides, consuming a larger quantity of these molecules in its transit to the pulp space. This coloring agent has been widely used in laboratory research to evaluate the bleaching efficacy and cytotoxicity of gels containing  $H_2O_2$ .  $^{12,14,15,19}$  Breakdown of the molecular structure of chromophores derived from black tea occurs gradually, so that part of the unreacted H<sub>2</sub>O<sub>2</sub> remains free to diffuse through tooth substrates, as previously observed in colorimetric analyses.  $^{12,14,15,19}$  This fact may explain the reduction in immediate cytotoxicity observed in this study, after we performed the bleaching procedure in pigmented discs (G4) in comparison with untreated bleached discs (G4). As reported by Sulieman,<sup>30</sup> the inorganic matrix present in many of the chromophores leads to them being trapped within the crystalline structure of the interprismatic region of enamel, which demands the local action of molecules with elevated oxidation potential to cause their fragmentation. As demonstrated in the literature, the decomposition of H<sub>2</sub>O<sub>2</sub> into different ROS with higher oxidation potential is relatively slow when H<sub>2</sub>O<sub>2</sub> is not catalyzed. 31,32 This makes reduction of the chroma (scale that refers to the quantity of saturation of the hue) occur in a gradual manner. 12,15,21 These data may perhaps explain, even if only partially, the fact that the in-office bleaching procedure assessed in this study had also caused an important cytotoxic effect, even when the procedure was used in teeth pigmented with black tea. We may suggest that the excess H<sub>2</sub>O<sub>2</sub> and its derivatives that did not interact with the pigmentation chromophores present in dental hard tissues ended up diffusing through this tubular substrate to cause damage to the pulp cells, such as we observed in the live/dead test.

In a recent study, Guo and others,  $^{23}$  validated the hypothesis that the rupture of organic chains, particularly those formed of aromatic compounds, arise from the process of oxidation promoted by  $H_2O_2$  and its decomposition byproducts. These oxidant agents, which are in a state of electronic instability, act directly on aromatic rings or in unsaturated linear carbon chains, reducing the rate of light absorption.  $^{33}$  It is important to point out that change in the color of the tooth structure results from

changes in the properties of light reflection, which are directly related to the incident light.<sup>34</sup> For this reason, it has been defined that the perception of light teeth after bleaching therapy is the result of the association of local chemical and physical phenomena. 9 As the tooth may present different bands of frequency in the emission of fluorescence within the visible light spectrum, 23 it is thus suggested that the behavior of H<sub>2</sub>O<sub>2</sub> may be muted by the chromophore on which this molecule acts. However, the aspects involving dental staining are complex because they may have intrinsic or extrinsic origin or even a combination of both,30 and this is a factor to be considered during bleaching therapies. Furthermore, there is a point of saturation in the dental structure that limits the bleaching potential of H<sub>2</sub>O<sub>2</sub> and consequently the tooth bleaching efficacy. 14-16 Therefore, one may suggest that, for each level of chromatic saturation of tooth, there is an ideal concentration of H<sub>2</sub>O<sub>2</sub> in the gel and an adequate time of application of the product on enamel to get the best esthetic outcome and to prevent the diffusion of high concentrations of this molecule capable of damaging pulp cells. In this way, dental bleaching appears to be a clinical procedure that should be increasingly individualized, so that the diffusion of residual H2O2 through dentin can be limited, preventing the negative side effects of this esthetic therapy.

As previously demonstrated in the literature,<sup>35</sup> excess intracellular H<sub>2</sub>O<sub>2</sub> interferes in the transcription factor peroxisome proliferator-activated receptor  $\gamma$  (PPAR $\gamma$ ), which acts in the removal of ROS during the inflammatory process, limiting the regenerative potential of tissues. 35 In these conditions, the expression of odontoblast differentiation markers, such as alkaline phosphatase (ALP), dentin sialophospho-protein (DSPP), dentin matrix phosphoprotein-1 (DMP-1), and deposition of mineralization nodules are intensely affected, which may harm the regeneration of injured tissues. 13-16 In the present research, we demonstrated the occurrence of pulp cell death due to necrosis caused by the intense cellular oxidative stress. We observed that the MDPC-23 cells were positively stained with the Eth-1 probe, which binds to the DNA of cells with disrupted membranes. Elevated concentrations of toxic reactive species result in direct cell death by necrosis<sup>36</sup>; in live tissues such as the pulp, this may cause the release of large quantities of intracellular components, triggering an acute local inflammatory reaction either associated with the formation of broad areas of necrosis or not. 2,37-39

In general, in the present laboratory study, it was possible to demonstrate that the presence of pigmentation in the tooth could reduce the diffusion of residual H<sub>2</sub>O<sub>2</sub>, and consequently, the trans-enamel and trans-dentin cytotoxicity of this reactive molecule. The enamel/dentin discs stained with black tea that were used in the present investigation to simulate human maxillary central incisors were obtained from bovine incisors. Because the bovine teeth have been considered adequate to replace human teeth, 40-45 a number of researchers have used this model to perform laboratory studies. 12-16 However, based on the fact that human teeth are more permeable to H<sub>2</sub>O<sub>2</sub> than bovine teeth, <sup>40</sup> one may suggest that the diffusion of this reactive oxygen species (ROS) through enamel/dentin and its cytotoxicity to pulp cells might be more intense for human teeth.

However, the data obtained in this investigation must be interpreted with caution, because the reduction in cytotoxicity did not occur with sufficient intensity to consider the tested bleaching treatment harmless to odontoblast cells. On the other hand, it could be inferred that the bleaching treatment in teeth with little chromatic saturation must be performed with mild doses, using products with low concentrations of peroxides. Furthermore, we emphasize that, for this study, only an experimental bleaching protocol was used, and this was the case for the intrinsic pigmentation protocol as well. Therefore, future studies must be conducted to investigate the different types of chromophores present in the dental substrate, as well as other bleaching therapies. Nevertheless, this study allowed for a greater biological understanding of the interrelations between pigmentation chromophores and H<sub>2</sub>O<sub>2</sub>, in a manner to allow collaboration with studies in the area of bleaching, particularly for refining laboratory studies in this field of esthetic dentistry.

#### **CONCLUSION**

It was concluded that, according to the methodology used in this investigation, the presence of pigments in hard tooth structures decreases the trans-enamel and trans-dentin diffusion of  $\rm H_2O_2$ , as well as the toxicity to pulp cells of an in-office bleaching gel with 35%  $\rm H_2O_2$ .

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#### **Regulatory Statement**

This study was conducted in accordance with all the provisions of the research ethics oversight committee guidelines and policies of the Univ Estadual Paulista–UNESP, Araraquara School of Dentistry.

#### **Conflict of Interest**

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Repair Surface Conditioning Measures Affect Enamel and Dentin Bond Strength

P Kanzow • L Piecha • J Biermann • A Wiegand

#### Clinical Relevance

Contamination of enamel and dentin with repair surface conditioning measures should be avoided.

#### **SUMMARY**

Objectives: To analyze whether the contamination with different repair conditioning measures impairs the adhesive performance of a universal adhesive applied in etch-and-rinse mode (ER) or self-etch mode (SE).

Methods and Materials: Bovine enamel and dentin surfaces (each subgroup n=16) were bonded with a universal adhesive in ER or SE after contamination with different repair conditioning measures (sandblasting, silica coating, hydrofluoric acid etching, self-etching

\*Philipp Kanzow, MSc, Dr med dent, Dr rer medic, Department of Preventive Dentistry, Periodontology and Cariology, University Medical Center Göttingen, Göttingen, Germany

Lisa Piecha, Department of Preventive Dentistry, Periodontology and Cariology, University Medical Center Göttingen, Göttingen, Germany

Jana Biermann, Dr med dent, Department of Preventive Dentistry, Periodontology and Cariology, University Medical Center Göttingen, Göttingen, Germany

Annette Wiegand, Prof Dr med dent, Department of Preventive Dentistry, Periodontology and Cariology, University of Göttingen, Göttingen, Germany

\*Corresponding author: Robert-Koch-Strasse 40, 37075 Göttingen, Germany; e-mail: philipp.kanzow@med. uni-goettingen.de

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ceramic primer). In half of the groups, sand-blasting, silica coating, and hydrofluoric acid etching was followed by the use of a universal primer. If the universal adhesive was applied in ER, contamination was performed either before or after phosphoric acid etching. If the universal adhesive was applied in SE, bonding was performed after contamination. In the control groups, no contamination was simulated. Shear bond strength (SBS) and failure modes of composite buildups were determined after thermal cycling (10,000 cycles, 5°C-55°C). Statistical analysis was performed using analyses of variance, Weibull statistics, and  $\chi^2$  tests (p<0.05).

Results: In ER, sandblasting and silica coating significantly reduced SBS (control: enamel =  $25.7\pm4.2$  MPa; dentin =  $22.0\pm5.3$  MPa) only when performed after phosphoric acid etching. Contamination with hydrofluoric acid impaired SBS on enamel but not on dentin. The self-etching ceramic primer reduced SBS, but not significantly. The contamination with the universal primer had no significant effect. In SE, all repair conditioning measures except the universal primer reduced SBS (control: enamel =  $20.3\pm5.5$  MPa; dentin =  $23.0\pm4.0$  MPa).

Conclusion: Contamination of enamel and dentin by repair conditioning measures potentially affects bond strength.

#### INTRODUCTION

Repair of partially defective dental restorations is becoming increasingly accepted by dentists and patients as it comes along with substantial advantages compared with complete replacement, for example, preservation of tooth structure, fewer pulpal complications, and reduced treatment time and costs.<sup>1</sup>

Optimal adhesion between the repair material (in most cases composite) and the original restoration makes complex conditioning of the repair surface necessary. Depending on the kind of restorative material, pretreatment of the repair surface to adhere to composite includes mechanical roughening, silica coating, sandblasting (composite, metal, zirconia), or hydrofluoric acid etching (glass ceramic), followed by the use of primers and adhesives containing silane/10-methacryloyloxydecyl dihydrogen phosphate (10-MDP).<sup>2,3</sup> However, in many clinical situations, the defective part of the restoration is surrounded by dental hard tissue, which makes the additional conditioning of enamel and/or dentin necessary. Especially in small cavities, contamination of enamel or dentin by repair conditioning measures is often unavoidable and might affect adhesion of composite to these dental hard tissues.

Several studies have shown that hydrofluoric acid application on dentin creates an amorphous layer of fluoride, rendering it difficult for adhesives to penetrate.4-7 Moreover, as hydrofluoric acid contamination also has the potential to cause chemical burn of surrounding soft tissues, alternative conditioning methods for intraoral repair of glass ceramic restorations are necessary. Recently, a self-etching glass ceramic primer containing ammonium polyfluoride and silane methacrylate was introduced to the market to replace conditioning with hydrofluoric acid and silanization. Although this product is not yet approved for intraoral use, it may be of interest if possible hazardous effects on hard and soft tissues can be avoided. So far, bonding performance of selfetching ceramic primer on lithium disilicate and feldspathic ceramics was shown to be limited compared with hydrofluoric acid etching.8,9 Nevertheless, bonding performance might still be acceptable for repaired restorations, especially if possible side effects are reduced. However, no information on contamination effects of the self-etching glass ceramic primer on dental hard tissues is so far available.

Besides hydrofluoric acid etching, potential side effects of silica coating and aluminum oxide sand-blasting were investigated but showed conflicting results, depending on the substrate (enamel or dentin) and kind of abrasion particle. <sup>10,11</sup>

However, potential detrimental effects of different repair conditioning measures and sequences have not been systematically analyzed, especially with regard to the bond strength of etch-and-rinse or selfetch adhesives. Therefore, in this study we aimed to analyze the shear bond strength of a universal adhesive applied in etch-and-rinse mode (ER) or self-etch mode (SE) on enamel and dentin after contamination with different repair surface conditioning measures and sequences. The null hypothesis tested was that the bond strength of the universal adhesive is not affected by the various repair conditioning measures.

#### **METHODS AND MATERIALS**

#### **Specimen Preparation**

Cylindrical enamel and dentin specimens (5.7 mm in diameter) were prepared from the crowns or roots of freshly extracted, nondamaged bovine incisors. The specimens were then embedded in chemically cured acrylic resin (Paladur, Kulzer, Hanau, Germany) and ground flat using sandpaper (WS flex 18C, grits 500 und 800, Hermes, Hamburg, Germany) while being water-cooled (RotoPol-35 and PdM-Force-20, Struers, Willich, Germany). Specimens were randomly divided into 25 groups of enamel or dentin specimens (n=16 for shear bond strength analysis, n=2 for scanning electron microscopy [SEM] analysis).

### Simulated Contamination and Bonding Procedures

The composition of the universal adhesive and the materials used in this study is presented in Table 1; surface contamination measures/sequences are shown in Tables 2 through 5.

The universal adhesive (Adhese Universal, Ivoclar Vivadent, Schaan, Liechtenstein) was either applied in ER (groups 1-16) or SE (groups 17-25) on enamel and dentin specimens.

If used in ER, 37% phosphoric acid was applied for 30 seconds (enamel) or 15 seconds (dentin) to the surface, rinsed with water for 30 seconds or 15 seconds (respectively) and gently air-dried. In the control group (group 1), the adhesive was applied without any simulated contamination and light-cured (20 seconds, B.A. Optima 10 LED, B.A. International, Hamburg, Germany). Contamination by sandblast-

Product	Manufacturer	Lot No	Composition	Application Mode
Adhese Universal	Ivoclar Vivadent, Schaan, Liechtenstein	W91987, X38352	HEMA (10%-25%), Bis-GMA (10%-25%), ethanol (10%-25%), 1,10-decandiol dimethacrylate ( $\geq$ 2.5%-<10%), methacrylated phosphoric acid ester ( $\geq$ 2.5%-<10%), campherquinone (1%-<2.5%), DMAEMA ( $\geq$ 1%-<2.5%)	20 s (scrubbing into the tooth surface using a disposable microbrush), 20 s light-curing
Filtek Supreme XTE	3M ESPE, Seefeld, Germany	N779140	Silane treated ceramic (60%-80%), silane treated silica (1%-10%), UDMA (1%-10%), bisphenol A polyethylene glycol diether dimethacrylate (1%-10%), Bis-GMA (1%-10%), silane treated zirconia (1%-5%), polyethylene glycol dimethacrylate (<5%), triethylene glycol dimethacrylate (<1%)	2-mm-thick increments, each 20 s light-cured
CoJet sand (silica- coated aluminum oxide)	3M ESPE, Seefeld, Germany	630975, 4236465	Aluminum oxide (>95%), synthetic amorphous silica (<5%)	4 s, 10-mm distance, 45°, 2-3 bar air pressure
Super Etch (phosphoric acid gel)	SDI, Bayswater, Australia	171138	Phosphoric acid (37%)	30 s (enamel) or 15 s (dentin), 30 s or 15 s (respectively) waterrinsing, gentle air-drying
Airsonic Alu-Oxyd (aluminum oxide)	Hager & Werken, Duisburg, Germany	N/A	N/A	4 s, 10-mm distance, 45°, 2-3 bar air pressure
Porcelain Etch (9% buffered hydrofluoric acid gel)	Ultradent Products, Cologne, Germany	BFTKV	Hydrofluoric acid (<10%)	90 s, water-rinsing (15 s)
Monobond Etch″	Ivoclar Vivadent, Schaan, Liechtenstein	X30890	Butanol (20%-<25%), tetrybutylammonium dihydrogen trifluoride (≤10%), methacrylated phosphoric acid ester (2.5%-<10%), bis(triethoxysilyl)ethane (1%-<2.5%)	20 s, after 40 s water-rinsing (15 s), gentle air-drying
Monobond Plus	Ivoclar Vivadent, Schaan, Liechtenstein	X11257	Ethanol (50%-100%), methacrylated phosphoric acid ester (1%-<2.5%)	20 s gentle air-blowing after 60 s

ing, silica coating, hydrofluoric acid etching, or the self-etching ceramic primer was performed either before (groups 10-16) or after (groups 3-9) phosphoric acid etching. In half of the groups, surfaces were also exposed to a universal primer.

If used in SE, the adhesive was applied for 20 seconds (enamel and dentin) before light-curing for 20 seconds (B.A. Optima 10 LED): In the respective control group (group 17), no contamination of the surfaces was performed. In the test groups, sand-blasting, silica coating, hydrofluoric acid etching, or the application of the self-etching primer was performed before the adhesive was applied. In half of the specimens in each group, surfaces were also contaminated with a universal primer.

The following surface contamination measures were applied either alone or in combination:

1) Air-abrasion with aluminum oxide (50  $\mu$ m; Hager & Werken, Duisburg, Germany) for 4 seconds at a distance of 10 mm (45°) and 2-3 bar air pressure. Remnants were air blown.

- 2) Silica coating: Surfaces were silica coated (30  $\mu$ m; CoJet, 3M ESPE, Seefeld, Germany) for 4 seconds at a distance of 10 mm (45°) and 2-3 bar air pressure. Loose particles were air blown.
- 3) Hydrofluoric acid (9% buffered hydrofluoric acid gel; Porcelain Etch, Ultradent Products, Cologne, Germany) was applied for 90 seconds. Samples were subsequently rinsed with water for 60 seconds.
- 4) The self-etching ceramic primer (Monobond Etch&Prime, Ivoclar Vivadent) was applied with a microbrush for 20 seconds followed by a 40-second contact time. Specimens were subsequently rinsed with water for 15 seconds.
- 5) Universal primer (Monobond Plus, Ivoclar Vivadent) was applied using a microbrush and allowed to evaporate for 60 seconds as recommended by the manufacturer. Afterward, samples were gently air blown.

After the universal adhesive was applied, the repair composite (Filtek Supreme XTE, 3M ESPE; shade A2) was adhered onto the specimen surface

using acrylic hollow cylinders (inner diameter: 3 mm, height: 4 mm). The composite was packed against the surface in a 2-mm-thick increment and light-cured for 20 seconds by applying the curing unit directly onto the acrylic cylinder. Light irradiance (>800 mW/cm²) was verified every 30 specimens using a radiometer (Cure Rite Model 644726, Dentsply Caulk, Milford, DE, USA). Contamination and bonding procedures were carried out by one operator (LP) throughout all experiments.

All specimens were then submitted to a thermal cycling procedure (10,000 cycles, between 5°C and 55°C, dwell time: 20 seconds, transfer time: 10 seconds) before shear bond testing.

## **Shear Bond Measurements and Failure Analysis**

Shear bond strength was tested with a universal testing machine (Materialprüfmaschine 1446, Zwick, Ulm, Germany). A shear force was applied to the adhesive interface through a chisel-shaped loading device at a crosshead speed of 1 mm/min parallel to the surface of the specimens. Load at fracture was recorded, and shear bond strength  $(\sigma)$  was calculated by software (TestXpert 11.02, Zwick) using the load at failure F(N) and the adhesive area  $A(mm^2)$ :  $\sigma = F/A$ .

The debonded area was examined for failure mode analysis with a stereomicroscope at 16× magnification (Stemi SV 11, Zeiss, Oberkochen, Germany). Failure mode was considered as adhesive if it occurred at the interface; as cohesive if at least parts of either enamel, dentin, or composite were affected; or as mixed when both adhesive and cohesive failures occurred.

#### **SEM Analysis**

Two specimens of each group were desiccated for 2 weeks in silica gel, sputter-coated with carbon, and analyzed by SEM (Ultra plus FE-SEM, Zeiss) at 10 kV.

#### **Statistical Analysis**

Statistical analyses were performed using the software SPSS Statistics for Macintosh (version 25.0.0.1, IBM, Armonk, NY, USA).

Separately for enamel and dentin, three-way analysis of variance (ANOVA) (ER without groups 9 and 16) and two-way ANOVA (SE without group 25) were performed. Factors were the sequence in which phosphoric acid etching was performed (first or second step, only in ER), the kind of contamination (sandblasting, silica coating, hydrofluoric acid

etching, control), and the use of a universal primer. Moreover, one-way ANOVAs followed by Scheffe post hoc tests were applied to compare all groups, including those where the self-etching ceramic primer was used.

Additionally, characteristic bond strength  $\sigma_0$  and Weibull modulus m (Weibull distribution parameters) were assessed using the maximum likelihood estimates and 95% confidence intervals in MATLAB (version R2018b, 9.5.0.1049112, The MathWorks, Natick, MA, USA).

For both substrates and each adhesive mode, effects of different repair surface treatments on failure modes were analyzed using  $\chi^2$  tests adjusted according to Bonferroni. The overall level of significance was set at  $\alpha = 0.05$ .

#### **RESULTS**

When the universal adhesive was applied in ER, shear bond strength values in enamel were significantly affected by the sequence of phosphoric acid etching (p<0.001, after contamination > before contamination) and the kind of contamination (p < 0.001, control > sandblasting = silica coating >hydrofluoric acid) but not by the contamination with the universal primer (p=0.056). The interaction between factors was not significant (p>0.169), except for the interaction between "sequence of phosphoric acid etching" and "kind of contamination" (p < 0.001). Between-group comparisons showed that groups 3-8, 14, and 15 led to significantly lower bond strength than the control. Shear bond strength, Weibull parameters, and failure modes of enamel specimens conditioned with the universal adhesive in ER are presented in Table 2.

If applied in SE, shear bond strength values on enamel were significantly affected by the kind of contamination (p<0.001, control) > sandblasting = silica coating > hydrofluoric acid) and by the use of a universal primer (p=0.028). The interaction between factors was not significant (p=0.389). Betweengroup comparisons showed that all repair contamination measures (except application of the universal primer, group 2) led to significantly reduced bond strength values. Shear bond strength, Weibull parameters, and failure modes of enamel specimens conditioned with the universal adhesive in SE are presented in Table 3.

On dentin, the shear bond strength of the universal adhesive applied in ER was significantly affected by the sequence of phosphoric acid etching (p<0.001, after contamination) > before contamina-

Table	2: Shear Bo	ond Strength, V	Veibull Parame	ters, and	Failure Modes	of Enamel Spe	ecimens in Etc	ch-and-rins	se Mod	e <sup>a</sup>
Group	Su	rface Contaminati	on/Conditioning		Bond Strength (MPa)	Weibull Param	eters [95% CI]	Faile	ure Mode	(%)
	1. Step	2. Step	3. Step	4. Step	Mean±SD	σ <sub>0</sub> (MPa)	m	Adhesive	Mixed	Cohesive
1	Phosphoric	None	- (Control)	Universal	25.7±4.2 ef	27.4 [25.5, 29.5]	7.2 [4.9, 10.7]	6.7	93.3	0.0
2	acid		Universal primer	adhesive	27.3±3.6 F	28.6 [27.4, 29.9]	11.6 [7.8, 17.3]	37.5	50.0	12.5
3	•	Sandblasting	_		15.8±4.3 ABC	17.4 [15.3, 19.8]	4.0 [2.8, 5.8]	75.0	25.0	0.0
4	•		Universal primer		16.0±1.7 ABC	16.8 [15.9, 17.6]	10.5 [7.2, 15.2]	56.3	43.8	0.0
5	•	Silica coating	-	='	11.3±3.6 A	12.5 [10.8, 14.6]	3.4 [2.4; 4.9]	50.0	50.0	0.0
6	•		Universal primer	='	12.6±2.3 A	13.6 [12.5, 14.7]	6.4 [4.3, 9.4]	31.3	68.8	0.0
7	•	Hydrofluoric	-	='	13.9±5.6 ab	15.6 [13.0, 18.7]	2.8 [1.9, 4.2]	25.0	68.8	6.3
8	•	acid	Universal primer	='	14.3±4.4 AB	15.9 [14.0, 18.1]	4.0 [2.6, 6.0]	25.0	75.0	0.0
9	•	Self-etching cera	mic primer		21.6±4.1 BCDEF	23.1 [21.5, 24.9]	6.8 [4.6, 10.2]	50.0	50.0	0.0
10	Sandblasting	Phosphoric acid	-	='	23.8±4.4 CDEF	25.4 [23.8, 27.0]	8.2 [5.4, 12.5]	43.8	43.8	12.5
11	•		Universal primer		23.9±5.7 CDEF	25.6 [23.5, 27.9]	5.9 [3.9, 8.9]	50.0	50.0	0.0
12	Silica coating	-	_	='	24.8±3.9 ef	26.4 [24.7, 28.2]	7.8 [5.3, 11.4]	0.0	92.9	7.1
13	•		Universal primer	='	24.5±3.7 DEF	26.2 [24.2, 28.2]	6.8 [4.8, 9.7]	12.5	87.5	0.0
14	Hydrofluoric	-	_	=	12.0±5.5 A	13.6 [11.0, 16.7]	2.5 [1.6, 3.7]	37.5	62.5	0.0
15	acid	_	Universal primer	- -	16.5±6.1 ABCD	18.5 [15.7, 21.6]	3.2 [2.1, 4.9]	25.0	68.8	6.3
16	Self-etching ceramic primer	-	-	-	18.9±6.4 ABCDE	20.9 [18.2, 23.9]	3.7 [2.4, 5.8]	46.7	46.7	6.7

<sup>&</sup>lt;sup>a</sup> Shear bond strength (mean $\pm$ standard deviation [SD], characteristic strength  $\sigma_0$  and Weibull modulus m with their 95% confidence intervals [Cls]) and failure modes of enamel specimens with the universal adhesive applied in etch-and-rinse mode. Different letters indicate significant differences between the groups as assessed by one-way analysis of variance followed by Scheffe post hoc tests.

tion) and the kind of contamination (p<0.001, control = hydrofluoric acid > sandblasting > silica coating) but not by the contamination with the universal primer (p=0.923). The interactions between factors were significant (p<0.043), except for the interaction between "sequence of phosphoric acid etching" and "kind of contamination" (p=0.268). Between-group comparisons revealed that bond strength was significantly reduced in groups 3-6 and 13 compared with the respective control. Shear bond strength, Weibull parameters, and failure modes of enamel specimens conditioned with the universal adhesive in ER are presented in Table 4.

If applied in SE, shear bond strength was affected by the kind of contamination (p<0.001, control > hydrofluoric acid > sandblasting > silica coating) and the use of the universal primer (p=0.001); the interaction between factors was not significant (p=0.210). Between-group comparisons showed that all contamination measures, except for the universal primer (group 18) and hydrofluoric acid/universal primer (group 24), led to significantly reduced bond strength. Shear bond strength, Weibull parameters, and failure modes of enamel specimens conditioned with the universal adhesive in SE are presented in Table 5.

Table 3	3: Shear Bond	Strength, Weibui	ll Paramete	ers, and Failure	Modes of Ename	l Specimens in	Self-etch l	Mode <sup>a</sup>	
Group	Surface Cont	amination/Condit	ioning	Bond Strength (MPa)	Weibull Parame	eters [95% CI]	Failure Mode (%)		
	1. Step	2. Step	3. Step	Mean±SD	σ <sub>0</sub> (MPa)	т	Adhesive	Mixed	Cohesive
17	None	- (Control)	Universal	20.3±5.5 c	22.3 [19.9, 24.9]	4.5 [3.1, 6.7]	31.3	43.8	25.0
18		Universal primer	adhesive	23.7±4.3 c	25.3 [23.6, 27.2]	7.3 [4.9, 10.9]	37.5	43.8	18.8
19	Sandblasting	-		11.4±5.3 в	12.8 [10.3, 15.9]	2.4 [1.6, 3.5]	73.3	26.7	0.0
20		Universal primer		13.6±3.9 в	14.9 [13.1, 16.9]	4.1 [2.8, 6.0]	60.0	40.0	0.0
21	Silica coating	-		12.8±3.8 в	14.1 [12.6, 15.8]	4.5 [2.9, 7.0]	64.3	35.7	0.0
22		Universal primer		13.8±3.4 в	15.1 [13.6, 16.7]	4.9 [3.3, 7.3]	80.0	20.0	0.0
23	Hydrofluoric acid	-		2.3±2.9 A	2.1 [1.2, 3.8]	0.9 [0.6, 1.3]	68.8	31.3	0.0
24		Universal primer		2.3±3.3 A	1.6 [0.6, 3.9]	0.5 [0.4, 0.8]	87.5	12.5	0.0
25	Self-etching ceran	nic primer	•	2.3±2.2 A	1.6 [0.5, 5.2]	0.4 [0.3, 0.7]	100.0	0.0	0.0

<sup>&</sup>lt;sup>a</sup> Shear bond strength (mean $\pm$ standard deviation [SD], characteristic strength  $\sigma_0$  and Weibull modulus m with their 95% confidence intervals [Cls]) and failure modes of enamel specimens with the universal adhesive applied in self-etch mode. Different letters indicate significant differences between groups as assessed by one-way analysis of variance followed by Scheffe post hoc tests.

Group	Surface	Contaminatio	n/Condition	ing	Bond Strength (MPa)	Weibull Param	eters [95% CI]	Failu	re Mod	e (%)
	1. Step	2. Step	3. Step	4. Step	Mean±SD	σ <sub>0</sub> (MPa)	т	Adhesive	Mixed	Cohesive
1	Phosphoric	None	- (Control)		22.0±5.3 E	24.0 [21.7, 26.6]	5.0 [3.4, 7.5]	53.3	46.7	0.0
2	acid		Universal primer	adhesive	21.2±3.8 E	22.7 [21.1, 24.4]	7.1 [4.8, 10.5]	25.0	75.0	0.0
3	•	Sandblasting	-	•	12.3±4.6 ABC	13.7 [11.6, 16.2]	3.0 [2.0, 4.5]	62.5	37.5	0.0
4			Universal primer	•	13.7±3.2 ABCD	14.9 [13.6, 16.3]	5.5 [3.7, 8.4]	68.8	31.3	0.0
5		Silica coating	_		9.6±3.0 AB	10.6 [9.3, 12.1]	3.9 [2.6, 5.8]	68.8	31.3	0.0
6			Universal primer	•	8.7±3.8 A	9.5 [7.4, 12.1]	2.0 [1.3, 3.1]	56.3	43.8	0.0
7		Hydrofluoric	-	•	15.6±5.3 ABCDE	17.0 [14.6, 19.8]	3.3 [2.1, 5.0]	43.8	56.3	0.0
8		acid	Universal primer		22.4±1.8 E	23.1 [22.4, 23.9]	15.9 [10.8, 23.6]	31.3	68.8	0.0
9		Self-etching ceramic prime	er	•	20.5±2.3 DE	21.5 [20.5, 22.5]	11.0 [7.5, 16.0]	43.8	56.3	0.0
10	Sandblasting	Phosphoric	_	•	16.2±4.4 BCDE	17.7 [15.9, 19.8]	4.7 [3.1, 7.1]	62.5	37.5	0.0
11		acid	Universal primer	•	17.6±3.1 CDE	18.7 [17.6, 19.8]	8.7 [5.8, 13.3]	64.3	35.7	0.0
12	Silica coating	_	_		17.9±5.3 CDE	19.7 [17.4, 22.4]	4.0 [2.7, 5.9]	28.6	71.4	0.0
13			Universal primer		10.0±4.2 AB	11.2 [ 9.2, 13.6]	2.7 [1.8, 4.0]	50.0	50.0	0.0
14	Hydrofluoric	_	_	•	20.7±3.2 DE	22.0 [20.6, 23.5]	7.7 [5.3, 11.3]	33.3	66.7	0.0
15	acid		Universal primer	•	22.0±5.3 E	23.9 [21.7, 26.4]	5.2 [3.5, 7.7]	18.8	81.3	0.0
16	Self-etching ceramic primer	-	_	•	17.6±4.5 CDE	19.3 [17.2, 21.6]	4.5 [3.1, 6.6]	87.5	12.5	0.0

<sup>&</sup>lt;sup>a</sup> Shear bond strength (mean $\pm$ standard deviation [SD], characteristic strength  $\sigma_0$  and Weibull modulus m with their 95% confidence intervals [Cls]) and failure modes of dentin specimens with the universal adhesive applied in etch-and-rinse mode. Different letters indicate significant differences between groups as assessed by one-way analysis of variance followed by Scheffe post hoc tests.

Highest Weibull parameters (characteristic strength  $\sigma_0$  and Weibull modulus m) were reached in the control groups or when solely the universal primer was applied, irrespective of the adhesive strategy and the substrate. The only exception was

found in the ER groups on dentin, where group 8 (phosphoric acid etching followed by hydrofluoric acid and the universal primer) obtained the highest Weibull modulus. For both enamel and dentin in ER, the majority of the groups reached higher charac-

Group	Surface Cont	amination/Condit	ioning	Bond Strength (MPa)	Weibull Parame	Weibull Parameters [95% CI]			e (%)
	1. Step	2. Step	3. Step	Mean±SD	σ <sub>0</sub> (MPa)	m	Adhesive	Mixed	Cohesive
17	None	- (Control)	Universal	23.0±4.0 DE	24.6 [23.0, 26.3]	7.6 [5.1, 11.5]	14.3	64.3	21.4
18		Universal primer	adhesive	24.9±4.6 E	26.7 [24.5, 29.2]	5.9 [4.1, 8.3]	0.0	28.6	71.4
19	Sandblasting	-		10.3±4.6 ab	10.1 [5.8, 17.7]	0.9 [0.5, 1.4]	86.7	13.3	0.0
20		Universal primer		12.3±2.3 ABC	13.2 [12.1, 14.5]	5.7 [4.0, 8.2]	53.3	46.7	0.0
21	Silica coating	-		7.3±4.0 A	8.2 [6.3, 10.7]	2.0 [1.3, 2.9]	85.7	14.3	0.0
22		Universal primer	•	8.1±4.9 AB	9.0 [6.6, 12.3]	1.6 [1.1, 2.5]	92.3	7.7	0.0
23	Hydrofluoric acid	-	•	13.1±5.1 вс	14.5 [12.2, 17.2]	2.9 [1.9, 4.5]	53.3	46.7	0.0
24		Universal primer	•	18.3±4.1 cD	19.9 [17.9, 22.1]	5.0 [3.4, 7.2]	46.2	53.8	0.0
25	Self-etching ceran	nic primer	•	9.6±4.1 AB	10.8 [8.9, 13.2]	2.6 [1.8, 3.8]	100.0	0.0	0.0

<sup>&</sup>lt;sup>a</sup> Shear bond strength (mean $\pm$  standard deviation [SD] characteristic strength  $\sigma_0$  and Weibull modulus m with their 95% confidence intervals [Cls]) and failure modes of dentin specimens with the universal adhesive applied in self-etch mode. Different letters indicate significant differences between groups as assessed by one-way analysis of variance followed by Scheffe post hoc tests.

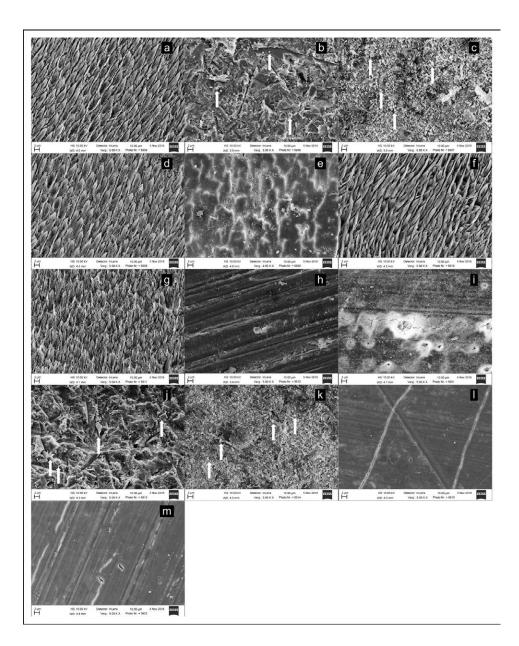


Figure 1. Representative scanning electron micrographs of enamel surfaces treated with sandblasting, silica coating, hydrofluoric acid etching, or the self-etching ceramic primer either alone (contamination before application of the adhesive in self-etch mode) or before/after phosphoric acid etching (contamination before application in etch-and-rinse mode). Note that contamination with the universal primer is not shown as it was not visible. Original magnification ×5000. (a): Phosphoric acid (control etch-andrinse, group 1). (b): Phosphoric acid → sandblasting (group 3). (c): Phosphoric acid → silica coating (group 5). (d): Phosphoric acid → hydrofluoric acid (group 7). (e): Phosphoric acid self-etching ceramic primer (group 9). (f): Sandblasting → phosphoric acid (group 10). (g): Silica coating phosphoric acid (group 12). (h): Hydrofluoric acid → phosphoric acid (group 14). (i): Self-etching ceramic primer → phosphoric acid (group 16). (j): Sandblasting (group 19). (k): Silica coating (group 21). (I): Hydrofluoric acid (group 23). (m): Self-etching ceramic primer (group 25). Abrasive remnants are marked by ↑.

teristic bond strengths when contamination was followed by phosphoric acid etching compared with phosphoric acid etching before contamination.

For both substrates and both adhesive application modes, the distribution of failure types differed significantly between different repair contaminations ( $p \le 0.003$ ).

#### **SEM Images**

SEM images are presented in Figures 1 (enamel) and 2 (dentin). Contamination of phosphoric acid etched enamel (Figure 1a) with sandblasting (Figure 1b) or silica coating (Figure 1c) resulted in removal of the etching pattern. When phosphoric acid etching was

performed after sandblasting (Figure 1f) or silica coating (Figure 1g), the surface presented an etching pattern not different from the control (Figure 1a). If sandblasting or silica coating, respectively, was not followed by phosphoric acid etching (ie, the universal adhesive was applied in SE), then the surface was distinctly roughened and remnants of abrasive particles could be seen (Figure 1j,k).

Hydrofluoric acid contamination of etched enamel resulted in an etching pattern with few visible surface precipitates (Figure 1d), while hydrofluoric acid etching of native enamel resulted in an amorphous surface layer (Figure 1h,l), which could not be removed by phosphoric acid etching (Figure 1h). Surface precipitation was also visible after

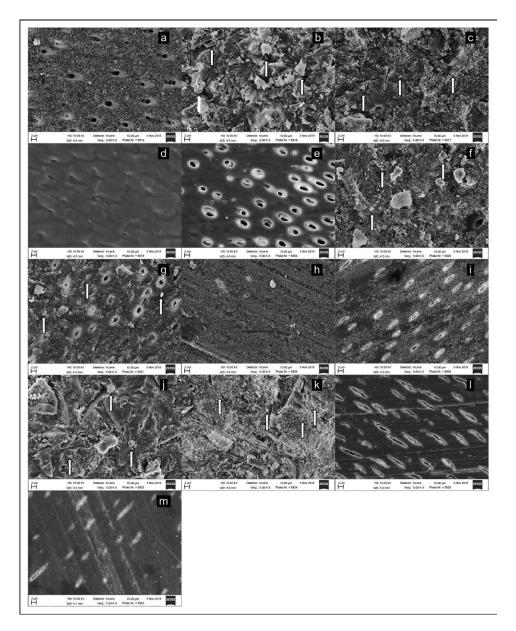


Figure 2. Representative scanning electron micrographs of dentin surfaces treated with sandblasting, silica coating, hydrofluoric acid etching, or the self-etching ceramic primer either alone (contamination before application of the adhesive in self-etch mode) or before/after phosphoric acid etching (contamination before application in etch-and-rinse mode). Note that contamination with the universal primer is not shown as it was not visible. Original magnification ×5000. (a): Phosphoric acid (control etch-andrinse, group 1). (b): Phosphoric acid → sandblasting (group 3). (c): Phosphoric acid → silica coating (group 5). (d): Phosphoric acid → hydrofluoric acid (group 7). (e): Phosphoric acid → self-etching ceramic primer (group 9). (f): Sandblasting → phosphoric acid (group 10). (g): Silica coating phosphoric acid (group 12). (h): Hydrofluoric acid → phosphoric acid (group 14). (i): Self-etching ceramic primer → phosphoric acid (group 16). (j): Sandblasting (group 19). (k): Silica coating (group 21). (I): Hydrofluoric acid (group 23). (m): Self-etching ceramic primer (group 25). Abrasive remnants are marked by ↑.

contamination with the self-etching ceramic primer (Figure 1e,i,m). Depending on the sequence of phosphoric acid etching, the surface precipitates either covered the enamel etching pattern (Figure 1e) or could not be removed by phosphoric acid etching (Figure 1i).

When etched dentin (Figure 2a) was contaminated by sandblasting (Figure 2b) or silica coating (Figure 2c), the surface was damaged and cracked, the exposed collagen network was removed, and dentinal tubules were no longer visible. When phosphoric acid etching was performed after sandblasting (Figure 2f) or silica coating (Figure 2g), the surface became partly etched and some tubules were visible. If sandblasting or silica coating was not followed by

phosphoric acid etching (ie, if the universal adhesive was applied in SE), the dentin surface was damaged (Figure 2j,k). Hydrofluoric acid contamination of etched dentin resulted in an amorphous surface precipitation (Figure 2d,h,l) that also partly covered the dentinal tubules. Slight surface precipitation was also visible after contamination with the self-etching ceramic primer (Figure 2e,i,m).

#### **DISCUSSION**

This study aimed to simulate potential contamination of dental hard tissues with different repair conditioning measures. Due to the large number of specimens, bovine specimens rather than human teeth were used. A recent systematic review found that bovine teeth can be a suitable alternative for human enamel and dentin substrates in shear bond strength tests as no significant differences have been detected in shear bond strength between human and bovine enamel and dentin. 12 Adhesion was tested using a universal adhesive containing 10-MDP, but without a silane component. A universal adhesive was chosen not only as it can be applied using different etching strategies (selective enamel etch, ER, and SE) but also as universal adhesives can be used for conditioning of restorative materials when restorations need to be repaired. The functional monomer 10-MDP can bond to zirconia and nonprecious metals by ionic and hydrogen interactions of the phosphate group with the oxide layer of the material<sup>13</sup> while silane-containing adhesives bond to silica-based materials or silica fillers by forming siloxane linkages. 14 Nevertheless, for repair conditioning of glass ceramics, a separate pretreatment step with a silane-containing solution or a universal primer is recommended to reduce the possibility of hydrolytic degradation of the ceramic-resin bonding. 15,16 As opposed to the universal adhesive, the universal primer includes not only phosphoric acid methacrylate but also silane methacrylate (capable of bonding to silicate surfaces) and disulfide methacrylate (capable of bonding to precious metals).

Besides the universal primer, the contamination effects of different repair measures to increase surface roughness of a restorative material by physical (sandblasting, silica coating) or chemical (hydrofluoric acid etching, self-etching ceramic primer) measures were tested. An increased surface roughness might allow for better wettability by the adhesive. In addition to surface roughening, silica coating might further improve repair bond strength due to the incorporation of silica particles. Thereby, formation of Si-O-Si-bonds to silane-containing adhesives or universal primers becomes possible. 17

For all simulated repair surface conditioning measures, the duration of contamination was based on the duration of application recommended for conventional use of the respective products. Under clinical conditions, contact time to dental hard tissues might be shorter, when unintended contamination is noticed and stopped.

For the ER, air abrasion and silica coating reduced enamel and dentin bond strength and increased the amount of adhesive failures when contamination was performed on etched surfaces. This observation is in line with previous studies showing reduced bond strength of etched enamel and dentin after silica coating. <sup>18,19</sup> The SEM images indicate that the

enamel etching pattern and the exposed collagen network of dentin was removed by sandblasting. Conversely, when phosphoric acid etching was performed after sandblasting contamination, bond strength values were not reduced and enamel and dentin specimens presented an etched surface. Previous studies indicated that sandblasting of dental hard tissues does not affect bond strength or margin quality adversely if sandblasted surfaces are cleaned with phosphoric acid 10,11 or water spray before adhesive application. For the SE, sandblasting contamination impaired bond strength values and increased the amount of adhesive failures, probably as the surface was not washed/cleaned, and loose abrasion particles were only air blown.

Hydrofluoric acid contamination of enamel resulted in significantly lower bond strength values, independently of the application sequence and the adhesive strategy. Saracoglu and others<sup>4</sup> demonstrated that 5% and 9.5% hydrofluoric acid inhibited enamel bond strength independently whether applied before or after phosphoric acid etching. However, in contrast to the present study, the etch-andrinse adhesive performed slightly better if hydrofluoric acid was applied after phosphoric acid etching instead of before.4 Interestingly, in the present study, enamel specimens presented clearly different surface characteristics (Figure 1d,h). While the sequence phosphoric acid etching/hydrofluoric acid contamination resulted in an etching pattern with only a few visible surface precipitates, the reverse order led to an amorphous surface without any signs of etching. Hydrofluoric acid is known to form dense amorphous fluoride precipitates on the tooth surface, which most probably cannot be removed by further phosphoric acid etching. However, irrespective of the sequence of phosphoric acid etching, hydrofluoric acid contamination significantly reduced enamel bond strength.

Shear bond strength measurement on hydrofluoric acid—exposed dentin yielded ambiguous results. Previous studies found that contact to hydrofluoric acid significantly impaired dentin bond strength, independent of the etching sequence. And the etching sequences is dentined differences between the etching sequences: If dentine was first etched with phosphoric acid, a collagen-rich layer was exposed, which probably collapsed when further etched with hydrofluoric acid. As a result, a thinner hybrid layer was formed and mineral precipitates were detected at the bottom of the hybrid layer. If hydrofluoric acid etching was performed first, calcium fluoride precipitates were

formed that hampered phosphoric acid etching. Underneath the precipitates, a collagen-rich layer was formed, which could not be infiltrated by resin.<sup>5</sup> In the present study, hydrofluoric acid contamination led to precipitates that were covering the surface (Figure 2d) or partial occlusion of dentinal tubules (Figure 21). Phosphoric acid etching of hydrofluoric acid contaminated dentin was unable to remove the surface precipitates (Figure 2h). Nevertheless, bond strength was significantly impaired only when the adhesive was applied in SE or when phosphoric acid etched dentin was contaminated (group 7). It remains unclear why the sequence hydrofluoric acid contamination/phosphoric acid etching led to only slightly reduced bond strength values, even after thermocycling. Similar results were obtained with the self-etching ceramic primer. On both enamel and dentin, the primer led to surface precipitates (Figure 1e,i,m, Figure 2e,i,m) that impaired bond strength significantly if the universal adhesive was applied in SE (Figures 1m and 2m). Etched enamel became visible underneath the precipitates (Figure 1i). Dentinal tubules were partly occluded by the precipitation (Figure 2e,i,m), but the precipitation seemed to be less severe than after hydrofluoric acid contamination. Further studies have to be performed to fully elucidate how the self-etching ceramic primer affects enamel and dentin adhesion.

The universal primer did not affect bond strength adversely. Previous studies showed that the contamination of enamel<sup>19</sup> or dentin<sup>21</sup> with silane solutions did not reduce bond strength of subsequently applied adhesives. Potentially, the universal primer increased the wettability of the contaminated enamel or dentin surface under certain conditions, leading to slightly higher bond strength values.

The brittleness of the materials included in this study makes their failure difficult to predict and their bond strength less reliable. The Weibull statistic takes into account the variable existence of strength-controlling flaws within brittle materials by providing information about the variability of their bond strengths. A high Weibull modulus reflects a narrow variability of bond strengths and consequently a high reliability on the characteristic strength. Materials or methods with a high Weibull modulus are likely to be less technique-sensitive than those with a low Weibull modulus and should therefore be preferred.<sup>22</sup> Irrespective of the substrate or the adhesive strategy, our results showed the highest Weibull moduli and characteristic strengths in the control groups or when only the universal primer was applied, with the exception of group 8. Thus, contamination of enamel or dentin by repair conditioning measures seemed to result in a less reliable compound between repair restoration and tooth structure and should therefore ideally be avoided.

The present study only assessed the effect of repair surface conditioning measures on enamel and dentin shear bond strengths. However, in a clinical setting mixed surfaces with both tooth and restoration surfaces require optimal repair pretreatment. The potential interference of tooth surface conditioning measures (ie, phosphoric acid etching) on repair bond strength was not assessed in the present study. In particular, zirconia and nonprecious metals should not be treated with phosphoric acid to avoid the formation of a stable phosphate layer inhibiting the adhesion of primers containing phosphoric acid methacrylate (eg, 10-MDP).<sup>23</sup>

Based on these results, the null hypothesis that repair conditioning measures do not affect bond strength of a universal adhesive to enamel or dentin has to be rejected. Within the limitations of this study, the following clinical recommendations for repairing restorations next to enamel and dentin substrates may be suggested: 1) contamination of enamel/dentin by repair conditioning measures should be avoided; 2) if enamel/dentin is contaminated, use of phosphoric acid (ER) before applying a universal adhesive may minimize the effects of the contamination; and 3) contamination of enamel/ dentin with a universal primer alone may not significantly affect the bond strength of dentin or enamel to resin composite when using a universal adhesive.

These recommendations are in line with a recently published policy statement on the repair of restorations published by the FDI World Dental Federation: When tooth substance is next to a restoration in need of repair, physical repair surface conditioning measures should be performed before phosphoric acid etching of the tooth substance.<sup>24</sup> However, it has also been suggested that repairs be performed in a two-step technique if tooth substance is present: In the first step, only the tooth surface is covered using composite. Afterward, physical/chemical repair surface conditioning measures are applied to both the composite and the adjacent restoration.<sup>25</sup> Given the results of the present study, this technique seems rational as it combines optimal repair bonding procedures for both tooth surfaces and restorations.

#### Note

This study was previously presented at the 9th CONSEURO, June 14-15, 2019, Berlin, Germany.

#### **Conflict of Interest**

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Effect of Layering Techniques on Polymerization Shrinkage Stress of High- and Low-viscosity Bulk-fill Resins

A Tsujimoto • CA Jurado • WW Barkmeier • ME Sayed T Takamizawa • MA Latta • M Miyazaki • F Garcia-Godoy

#### Clinical Relevance

The use of layering techniques is still advisable with many bulk-fill resins and should be the default unless a particular resin is known to not need it.

#### **SUMMARY**

Objective: The purpose of this study was to investigate how layering techniques affect polymerization shrinkage stresses of high-and low-viscosity bulk-fill resins.

Method: Six high-viscosity and six low-viscosity bulk-fill resins were evaluated. Aluminum blocks with a mesial-occlusal-distal (MOD) cavity were machined and randomly divided into groups for different filling techniques (bulk-fill vs horizontal layering vs oblique layering) and further subdivided according to

Toshiki Takamizawa, DDS. PhD, Operative Dentistry, Nihon University School of Dentistry, Tokyo, Japan type of resin (high- vs low-viscosity). The cuspal deflection resulting from the polymerization of bulk-fill resin bonded to a MOD cavity within an aluminum block was measured with a digimatic micrometer. Scanning electron microscopy analyses of tested resins were also conducted.

Results: In the high-viscosity bulk-fill resins, cuspal deflection of the MOD cavity ranged from 11.2 to 18.2  $\mu m$  with the bulk-filling technique, from 10.7 to 15.5  $\mu m$  with the horizontal layering technique, and from 10.9 to 15.2  $\mu m$  with the oblique layering technique. In the

Mark A Latta, DMD, MS, General Dentistry, Creighton University School of Dentistry, Omaha, NE, USA

Masashi Miyazaki, DDS, PhD, Operative Dentistry, Nihon University School of Dentistry, Tokyo, Japan

Franklin Garcia-Godoy, DDS, MS, PhD, PhD, Bioscience Research, University of Tennessee Health Science Center College of Dentistry, Memphis, TN, USA

\*Corresponding author: 1-8-13, Kanda-surugadai, Chiyoda-ku, Tokyo, 101-8310, Japan; e-mail: tsujimoto.akimasa@nihon-u. ac.jp

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<sup>\*</sup>Akimasa Tsujimoto, DDS, PhD, Operative Dentistry, Nihon University School of Dentistry, Tokyo, Japan

Carlos A Jurado, DDS, MS, Clinical Digital Dentistry, A.T. Still University Arizona School of Dentistry and Oral Health, Mesa, AZ, USA

Wayne W Barkmeier, DDS, MS, General Dentistry, Creighton University School of Dentistry, Omaha, NE, USA

Mohammed E Sayed, BDS, MDS, FRCDC, FACP, AFAAID, PhD, Prosthodontics, Jazan University College of Dentistry, Jazan, Saudi Arabia

low-viscosity bulk-fill resins, cuspal deflection of the material ranged from 9.2 to 19.8  $\mu m$  with the bulk-filling technique, from 8.2 to 15.7  $\mu m$  with the horizontal layering technique, and from 8.4 to 16.4  $\mu m$  with the oblique layering technique.

Conclusion: Cuspal deflections for some highand low-viscosity bulk-fill resins were significantly reduced by using layering techniques, but the resultant improvement of layering techniques was not applicable to all the bulkfill resins used in this study.

#### **INTRODUCTION**

Restorative procedures using composite resin have become an essential part of daily practice because of material enhancements<sup>1</sup> and improvements in adhesive systems.<sup>2</sup> Material improvements over the years have resulted in an increase in patient demands for esthetics<sup>3</sup> and more emphasis on preserving tooth structure.4 The wear resistance of composite resins has continued to improve, since their introduction as restorative materials, enabling expanded use in posterior restorations of large cavities with long-term durability.<sup>5</sup> A systematic review<sup>6</sup> comparing direct and indirect composite resin restorations in posterior teeth showed that there was no difference in longevity between direct and indirect restorations, regardless of the type of composite resin. In addition, a recent 20-year clinical study of direct composite resin restorations in posterior teeth showed a high success rate and increased mean survival time.

However, polymerization shrinkage and its associated stress have remained a major concern of direct resin restorations, especially in large cavities in posterior teeth.8 Numerous studies have been performed to assess polymerization shrinkage and stress development in composite resins. 9,10 Polymerization shrinkage causes stress at the interface between a tooth and a restoration as the elastic modulus of the resin increases during polymerization. 11 This stress may result in marginal gap formation, microleakage, and enamel micro-cracks and can give rise to pulpal irritation, secondary caries due to bacterial infiltration, and postoperative sensitivity, which in turn can lead to restoration failure requiring restoration replacement (Figure 1). 12 Efforts have been made to reduce polymerization shrinkage and stress by increasing inorganic filler loading, 13 modifying filler particle size and shape, 14 and developing new types of resin matri- $\cos^{15}$ 

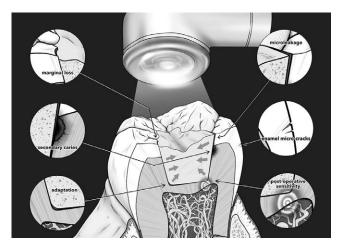


Figure 1. Schematic drawing of the influence of polymerization shrinkage stress.

However, increases in inorganic filler loading and modification of filler size and shape are limited by the need for a physicochemical combination within the resin matrix. Polymerization shrinkage and stress of composite resins can also be reduced by decreasing the reactive sites per unit volume through increasing the molecular weight per reactive group. 16 Nevertheless, this strategy also has its limitations because the use of monomers with a high molecular weight may produce increased viscosity, resulting in poorer handling characteristics, and potentially reducing mechanical strength. In addition, attempts to develop composite resins with completely new resin matrices that shrink considerably less, have been ongoing. Recently, a composite resin with a silorane resin matrix containing siloxane and oxirane was introduced, but this resin needs special adhesive systems for bonding.<sup>17</sup>

Clinical strategies to minimize the shrinkage stress of composite resins, such as horizontal and oblique layering techniques to decrease the C-factor, are still widely used. 18 In addition, recent developments have led to the introduction of high- and lowviscosity bulk-fill resins that can be used for large cavities with the bulk-fill technique. 19 These materials are designed to be used for placing the resin in increments of up to 4 mm, and one of their central characteristics is a much greater depth of cure than that found in conventional resins.<sup>20</sup> However, they have also been designed to reduce polymerization shrinkage stress to support their use in bulk-fill techniques.<sup>5</sup> No independent research has yet investigated whether the filling techniques used to reduce polymerization shrinkage stress are still necessary or valuable with these newly developed bulk-fill resins.

Although measurement of cuspal deflection is a useful way to evaluate the polymerization shrinkage stress of composite resins, the use of extracted teeth to measure cuspal deflection can produce significant discrepancies among specimens due to the lack of standardization of the anatomical and histochemical characteristics of the individual teeth.<sup>21</sup> Recently, Tsujimoto and others<sup>5</sup> fabricated aluminum blocks with identical shapes and dimensions instead of using extracted human teeth to examine cuspal deflection. The cuspal deflection of the aluminum blocks was measured using a digimatic micrometer or a confocal laser scanning microscope.

This laboratory study examined cuspal deflection of bulk-fill resins using standardized uniform aluminum blocks with mesial-occlusal-distal (MOD) cavity preparations. The purpose of this study was to investigate the influence of filling techniques on the polymerization shrinkage stresses of high- and low-viscosity bulk-fill resins. The null hypothesis was that the layering techniques would have no influence on the polymerization shrinkage stresses of high- and low-viscosity bulk-fill resins.

#### **METHODS AND MATERIALS**

#### **Study Materials**

Six high-viscosity bulk-fill resins were used: 1) Beautifil Bulk (BB, Shofu, Kyoto, Japan); 2) EverX Posterior (EP, GC, Tokyo, Japan); 3) Filtek Bulk Fill (FB, 3M OralCare, St Paul, MN, USA); 4) Quixx Fill Posterior Restorative (QF, Dentsply Sirona, York, PA, USA); 5) Tetric Evo Ceram Bulk Fill (TEB, Ivoclar Vivadent, Schaan, Liechtenstein); and 6) Tetric N Ceram Bulk Fill (TNB, Ivoclar Vivadent). Six low-viscosity bulk-fill resins were used: 1) Beautifil Bulk Flow (BF, Shofu); 2) EverX Flow (EF, GC); 3) Filtek Fill and Core Flowable Restorative (FF, 3M Oral Care); 4) SDR Flow+ (SD, Dentsply Sirona); 5) Tetric Evo Flow Bulk Fill (TF, Ivoclar Vivadent), and 6) X-tra Base (XB, Voco GmbH, Cuxhaven, Germany).

#### **Specimen Preparation**

Aluminum blocks (10 [W]  $\times$  8 [L]  $\times$  15 [D] mm) with a simulated MOD cavity (4 [W]  $\times$  8 [L]  $\times$  4 [D] mm) were milled out using a computer-aided design/computer-aided manufacturing system, creating two remaining cusps. This structure simulated a large Class II cavity preparation in a premolar. The inside of the cavity was submitted to airborne-particle abrasion with 50  $\mu m$   $Al_2O_3$  powder for 10 seconds to improve surface characteristics for bond-

ing. Air pressure was set to 0.2 MPa, and the distance between the tip and aluminum surface was approximately 10 mm (Jet Blast II, J. Morita, Tokyo, Japan). A universal adhesive (Scotchbond Universal Adhesive, 3M Oral Care) was applied according to the manufacturer's instructions before placing the high- and low-viscosity bulk-fill resins. The adhesive was light-cured for 10 seconds at a standardized distance of 1 mm using a quartz–tungsten–halogen curing unit (OptiLux 501, Kerr, Orange, CA, USA). The power density (>700 mW/cm²) of the curing unit was confirmed with a dental radiometer (Model 100, Kerr) before specimen preparation.

#### **Measurement of Cuspal Deflection**

The aluminum blocks were randomly divided into three groups for different filling techniques (bulk-fill vs horizontal layering vs oblique layering technique) and were further subdivided according to the type of composite resin (high- vs low-viscosity) (Figure 2).

Group 1 (Bulk-fill Technique)—High- or low-viscosity bulk-fill resin was placed in bulk and light-cured for 40 seconds for each of the three exposed surfaces. Cuspal deflection was measured from the difference in the distance between the center of the two remaining cusps before the placement of composite resin and 10 minutes after light curing using a high-accuracy submicron digimatic micrometer (MDH-25MB, Mitutoyo, Tokyo, Japan).

Group 2 (Horizontal Layering Technique)—Highor low-viscosity bulk-fill resin was placed in two separate horizontal increments (2 mm each). Each increment was light-cured for 40 seconds each to the three exposed surfaces to ensure that an identical curing time was maintained. Cuspal deflection was measured in the same manner as in group 1.

Group 3 (Oblique Layering Technique)—High- or low-viscosity bulk-fill resin was placed in three separate oblique increments. Each increment was light-cured for 40 seconds each to the three exposed surfaces to ensure that an identical curing time was maintained. Cuspal deflection was measured in the same manner as in group 1 and group 2.

#### Scanning Electron Microscopy (SEM) Observations

SEM (TM 3000, Hitachi-High Technology, Tokyo, Japan) was used to examine the filler size and shape of the tested high- and low-viscosity bulk-fill resins. A thin coating of gold-palladium alloy was applied in a sputter coater (SC7620 Mini Sputter Coater, Emitech, East Sussex, UK) to prevent electrostatic

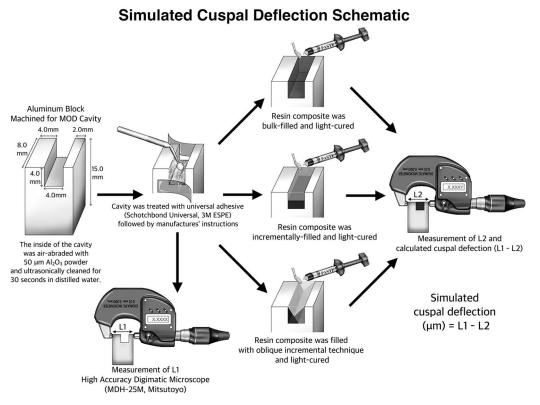


Figure 2. Schematic drawing of the experimental set-up for simulated cuspal deflection of bulk-fill resins.

charge from accumulating at the sample surface. The observations were done at an operating voltage of 15 kV.

#### **Statistical Analysis**

Statistical analysis was conducted with a commercial statistical software package (SPSS Statistics 25, IBM; Armonk, NY, USA). Two-way analysis of variance with a Tukey post hoc honest significant difference test was used to analyze the gathered data for the influence of 1) layering technique and 2) type of resin with an  $\alpha$  level of 0.05.

#### **RESULTS**

#### **Cuspal Deflection**

The effect of layering techniques on polymerization shrinkage stresses is shown in Table 2 for high-viscosity bulk-fill resins and in Table 3 for low-viscosity bulk-fill resins. Cuspal deflection for the high-viscosity resins ranged from 11.2 to 18.2  $\mu m$  with the bulk-fill technique, 10.7 to 15.5  $\mu m$  with the horizontal layering technique, and 10.9 to 15.2  $\mu m$  with the oblique layering technique. The rank order of cuspal deflection using the bulk-filling technique was TEB-FB-TNB-QF-EP-BB and that

using the layering technique was TEB-EP-FB-TNB-QF-BB.

Cuspal deflection for the low-viscosity resins ranged from 9.2 to 19.8  $\mu m$  with the bulk-fill technique, 8.2 to 15.7  $\mu m$  with the horizontal layering technique and 8.4 to 16.4 with the oblique layering technique. The rank order of cuspal deflection using the bulk-filling technique was SD-FF-TF-EF-XB-BF and that using the layering techniques was SD-FF-EF-TF-XB-BF.

The cuspal deflection of BB, EP, and QF in highviscosity resins and the cuspal deflection of BF, EF, and XB in low-viscosity resins were decreased when the layering techniques were used, unlike those of other resins.

#### **SEM Observation**

Representative SEM observations of tested resins are shown in Figure 3. The SEM images clearly show that there are differences in the size and shape of fillers in the high- and low-viscosity bulk-fill resins. BB, TEB, TNB, and QF among high-viscosity resins as well as BF, SD, TF, and XB among low-viscosity resins showed irregular particles with a broad size range (<1 to 20  $\mu m$ ). EP and EF showed short E glass fibers of different diameters and irregular

Material Type	Material (Code)	Matrix Resin Composition	Filler Composition	Manufacturer
High-viscosity bulk-fill resin	Beautifil Bulk (BB)	Bis-GMA, Bis-MPEPP, UDMA, TEGDMA	Fluoro-silicate glass	Shofu, Kyoto, Japan
	EverX Posterior (EP)	Bis-GMA, PMMA, TEGDMA	Short E-glass fiber filler, barium glass	GC, Tokyo, Japan
	Filtek Bulk Fill (FB)	Bis-EMA, Bis-GMA, TEGDMA	Silica filler, zirconia filler, zirconia/silica cluster filler	3M Oral Care, St Paul, MN, USA
	Quixx Fill Posterior Restorative (QF)	Bis-EMA, UDMA, TEGDMA	Strontium-aluminum- sodium-fluoridephosphate- silicate glass	Dentsply Sirona, York, PA, USA
	Tetric Evo Ceram Bulk Fill (TEB)	Bis-EMA, Bis-GMA, UDMA	Silanated barium glass filler	Ivoclar Vivadent, Schaan, Liechtenstein
	Tetric N Ceram Bulk Fill (TNB)	Bis-EMA, Bis-GMA, UDMA	Silanated barium glass filler	Ivoclar Vivadent
Low-viscosity bulk-fill resin	Beautifil Bulk Flowable (BF)	Bis-GMA, Bis-MPEPP, TEGDMA, UDMA	Fluoro-silicate glass	Shofu
	Ever X Flow (EF)	Bis-GMA, PMMA, TEGDMA	Short E-glass fiber filler, barium glass	GC
	Filtek Fill and Core Flow (FF)	Bis-GMA, UDMA	Silica filler, zirconia filler, zirconia/silica cluster filler	3M Oral Care
	SDR Flow+ (SD)	Bis-EMA, modified TEGDMA, UDMA	Barium-fluoro-alumino- silicate glass, strontium- fluoro-alumino-silicate glass	Dentsply Sirona
	Tetrik Evo Bulk Flow (TF)	Bis-EMA, Bis-GMA, UDMA	Silanated barium glass filler	Ivoclar Vivadent
	X-tra Base (XB)	Aliphatic dimethacrylate, Bis- EMA	Inorganic fillers	Voco GmbH, Cuxhaver Germany

methacryloxypolyethoxyphenyl)propane; PMMA, polymethyl methacrylate; TEGDMA, triethylene glycol dimethacrylate; UDMA, urethane dimethacrylate.

particles of a relatively uniform small size ( $<1~\mu m$ ). The concentration of short E glass fibers in EF was higher than that in EP. FB and FF showed quite small ( $<1~to~5~\mu m$ ) irregular, and spherical particles.

#### **DISCUSSION**

Cuspal deflection is one of the accepted indicators of polymerization shrinkage stress of composite resins

and has been evaluated using devices such as linear variable differential transformers, direct current differential transformers, and strain gauges. Recently, Tsujimoto and others developed a cuspal deflection measurement technique using a digimatic micrometer. Compared with previously reported methods, the digimatic micrometer may be a more accessible and practical approach to measuring cuspal deflection than use of confocal scanning laser

High-viscosity Bulk-fill Resin (Code)		Cuspal Deflection (µm)	
	Bulk-fill	Horizontal Layering	Oblique Layering
Beautifil Bulk (BB)	17.2 (0.6) aA	15.5 (0.8) aB	15.2 (0.7) aB
EverX Posterior (EP)	18.2 (0.8) aA	11.9 (0.9) bB	12.7 (0.8) bB
Filtek Bulk Fill (FB)	13.3 (0.9) bA	12.3 (0.9) bA	12.8 (0.8) bA
Quixx Fill Posterior Restorative (QF)	14.8 (0.5) cA	14.2 (0.6) cA	14.1 (0.6) cA
Tetric Evo Ceram Bulk Fill (TEB)	11.2 (0.6) dA	10.7 (0.7) dA	10.9 (0.6) dA
Tetric N Ceram Bulk Fill (TNB)	13.5 (0.9) bA	12.5 (1.2) bA	12.6 (1.1) bA

<sup>&</sup>lt;sup>a</sup> Values in parenthesis are standard deviations (n=10). The same lowercase letter in the same vertical column indicates no significant difference (p>0.05). The same uppercase letter within an individual row indicates no significant difference (p>0.05).

Table 3: Cuspal Deflection of Low-viscosity Bulk-fill Resins <sup>a</sup>							
	Cuspal Deflection (μm)						
Bulk-fill	Horizontal Layering	Oblique Layering					
19.8 (1.0) aA	15.7 (0.9) aB	16.3 (0.9) aA					
16.3 (1.1) bA	12.1 (0.8) bA	12.3 (0.9) bA					
11.0 (0.9) cA	10.1 (1.0) cA	10.4 (0.8) cA					
9.2 (0.8) dA	8.2 (1.1) dA	8.4 (1.2) dA					
14.2 (0.9) eA	13.7 (0.8) eA	13.6 (0.9) eA					
17.3 (0.7) bA	14.4 (0.8) eB	14.2 (0.9) eB					
	Bulk-fill 19.8 (1.0) aA 16.3 (1.1) bA 11.0 (0.9) cA 9.2 (0.8) dA 14.2 (0.9) eA	Cuspal Deflection (μm)Bulk-fillHorizontal Layering19.8 (1.0) aA15.7 (0.9) aB16.3 (1.1) bA12.1 (0.8) bA11.0 (0.9) cA10.1 (1.0) cA9.2 (0.8) dA8.2 (1.1) dA14.2 (0.9) eA13.7 (0.8) eA					

<sup>&</sup>lt;sup>a</sup> Values in parenthesis are standard deviations (n=10). The same lowercase letter in the same vertical column indicates no significant difference (p>0.05). The same uppercase letter within an individual row indicates no significant difference (p>0.05).

microscopy and other devices. In this study, a digimatic micrometer was used to measure cuspal deflection for high- and low-viscosity bulk-fill resins.

Park and others<sup>23</sup> reported results that the cuspal deflection in the layering technique was shown to be considerably lower than that for the bulk-fill technique, and they found no significant difference between horizontal and oblique layering techniques using a conventional resin. However, in that study, the resin used for the bulk-fill technique was a

conventional resin with a recommended maximum 2-mm depth of cure. Therefore, the depth of polymerization might not be totally completed in the bulk-fill specimens, making it more difficult to interpret test results. Further, as the bulk-fill technique is not recommended for clinical use with conventional resins, the clinical application of their results may not be applicable.

Recently, high- and low-viscosity bulk-fill resins have been used to expedite the restoration process by

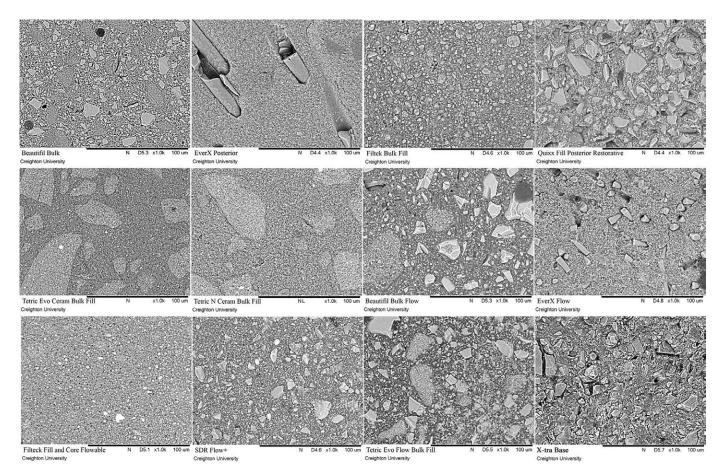


Figure 3. Scanning electron microscopy observations of the surfaces of high- and low-viscosity bulk-fill resins.

enabling increments up to 4-mm thick to be lightcured. <sup>19,20</sup> The present study compared the influence of layering techniques on high- and low-viscosity bulk-fill resins. Bulk-fill resins face no problems with polymerization in smaller increments to reduce polymerization shrinkage stress and can be used in bulk-fill or layering techniques in the clinic, as appropriate to the situation. Thus, it is clinically important to determine whether there are reasons to continue to use layering techniques with bulk-fill resins. While Park and others<sup>23</sup> found a significant difference in conventional resin composites, it is not surprising that the present study showed different behavior in cuspal deflection for bulk-fill resins.

The 12 bulk-fill resins used in this study can be classified into four groups according to the level of shrinkage stress produced: 1) high-viscosity bulk-fill resins with low stress (TEB, FB, QP, and TNB); 2) high-viscosity bulk-fill resins with high stress (BB and EP); 3) low-viscosity bulk-fill resins with low stress (SD, FF, and BB); and 4) low-viscosity bulk-fill resins with high stress (BF, EF, and XB). A highviscosity bulk-fill resin (BB) and a low-viscosity bulk-fill resin (SF) showed the highest and lowest values of cuspal deflections, respectively. Although, within the high- and low-viscosity bulk-fill resins with low stress, there was no significant difference (p>0.05) between the value of the cuspal deflection measured with bulk-filling and that measured with layering techniques. For high- and low-viscosity bulk-fill resins with high stress, the difference in cuspal deflection was significant (p < 0.05). This suggests that, for low shrinkage stress materials, advances in material composition are more important for reducing polymerization shrinkage stress than choice of filling techniques. However, this cannot be said of materials that show high polymerization shrinkage stress.

It is generally accepted that horizontal and oblique layering techniques improve marginal adaptation and bond stability of the adhesive interface between tooth structures and resin. <sup>24</sup> Therefore, when filling a large cavity, the use of small increments horizontally and obliquely was still recommended in one of the reviews <sup>25</sup> so that the polymerization shrinkage stress could be reduced. Although it is true that all the tested materials showed decreased values of cuspal deflection in this study with horizontal and oblique layering techniques, the values did not show statistically significant differences in high- and low-viscosity bulk-fill resins with low stress. Therefore, it appears that the use of polymerization shrinkage reduction technology in resins is more important

than layering techniques. This is consistent with recent systematic reviews. <sup>26,27</sup> In those reviews, the technique protocols of placing material no longer showed a great contribution to polymerization shrinkage stress and, indeed, made less difference than the choice of light-curing method. On the other hand, modifying the resin matrix made the largest contribution to minimizing polymerization shrinkage stress development.

The layering technique for composite resins increases treatment time and increases the risk of creating voids and/or other weaknesses within a restoration.<sup>28</sup> Thus, in low polymerization shrinkage bulk-fill resins, the rationale for using layering techniques is diminished because the layering technique has a minimal influence on polymerization shrinkage stress. However, this was not true for all of the bulk-fill resins investigated in the present study; thus, clinicians need to be familiar with the properties of the bulk-fill resin in order to choose the most appropriate procedures. Hayashi and others<sup>29</sup> have shown that bulk-fill resins with higher rates of polymerization shrinkage may nevertheless show much lower gap formation than other resins. This suggests that total polymerization shrinkage and stress are not enough information to guide selection of an appropriate technique. and that detailed information about the interaction between resins and adhesives is necessary. As this information is not easily available outside the research literature, the results of this study suggest that at present it would be safer to continue to use the layering technique as a significant number of bulk-fill resins show significantly higher polymerization shrinkage stress without it. Thus, from the overall results of this study, the null hypothesis that the layering method would have no influence on polymerization shrinkage stress, was rejected for some materials but not for all.

#### CONCLUSIONS

The results of this study indicate that the effect of layering techniques on polymerization shrinkage stress for high- and low-viscosity bulk-fill resins was material dependent. A decrease in polymerization shrinkage stress with layering techniques was not observed in bulk-fill resins that showed low cuspal deflection ( $<15~\mu m$ ).

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#### Conflict of Interest

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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# Interfacial Evaluation of CAD/CAM Resin Inlays on the Cavity Floor Using Swept-source Optical Coherence Tomography

S-H Han • Y Shimada • A Sadr • J Tagami • S-E Yang

#### Clinical Relevance

When a resin nanoceramic inlay is cemented using self-adhesive cement, a universal dentin adhesive can be applied to the prepared cavity. The application of the adhesive before self-adhesive cement placement provides similar or better interfacial adaptation than without the adhesive.

#### **SUMMARY**

#### Purpose: The first objective of this study was to determine whether the luting material used

Seung-Hoon Han, DDS, PhD, assistant professor, Department of Conservative Dentistry, St. Vincent Hospital, College of Medicine, The Catholic University of Korea, Gyeonggi-do, South Korea

Yasushi Shimada, DDS, PhD, associate professor, Department of Operative Dentistry, Okayama University, Okayama, Japan; and Department of Cariology and Operative Dentistry, Tokyo Medical and Dental University, Tokyo, Japan

Alireza Sadr, DDS, PhD, associate professor, Department of Restorative Dentistry, School of Dentistry, University of Washington, Seattle, Washington, USA

Junji Tagami, DDS, PhD, professor, Department of Cariology and Operative Dentistry, Tokyo Medical and Dental University, Tokyo, Japan

\*Sung-Eun Yang, DDS, PhD, professor, Department of Conservative Dentistry, Seoul St. Mary's Hospital, College of Medicine, The Catholic University of Korea, Seoul, Korea

\*Corresponding author: 222, Banpo-daero, Seocho-gu, Seoul 06591, Korea; e-mail: dentyeun@catholic.ac.kr

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for computer-aided design and computer-aided manufacture resin nanoceramic inlays affected interfacial adaptation. The second objective was to investigate whether application of a universal dentin adhesive before cementation affected interfacial adaptation. The final objective was to compare the inlay-side and dentin-side interfaces in the cement space.

Methods and Materials: Seventy-four class I cavities were prepared on extracted human third molars. Cavities were optically scanned, and resin nanoceramic inlays were milled using Lava Ultimate blocks (3M ESPE). For the control groups, the fabricated inlays were cemented using Panavia V5 (Kuraray Noritake) or FujiCem 2 (GC). For the experimental groups, the teeth were randomly divided into groups I and II. Group I contained four subgroups using different luting materials; in all subgroups, the inlays were cemented and dual cured without pretreatment. Group II contained six subgroups in which inlays were cemented and dual cured after application of

a universal dentin adhesive. After thermocycling, interfacial adaptation was measured using swept-source optical coherence tomography (SS-OCT) imaging and statistically compared among groups.

Results: Interfacial adaptation was different depending on the luting material used (p<0.05). After application of a universal adhesive, some subgroups showed improved interfacial adaptation (p<0.05). In the comparison of inlay-side and dentin-side interfaces, no difference was found in interfacial adaptation (p>0.05).

Conclusions: Interfacial adaptation for resin nanoceramic inlays differed with luting material. For some self-adhesive cements, application of a universal adhesive before cementation improved interfacial adaptation.

#### INTRODUCTION

Computer-aided design and computer-aided manufacturing (CAD/CAM) restorations, using prefabricated blocks, is one of the latest trends in restorative dentistry. As well as ceramic materials, indirect composite inlays can be milled from prefabricated composite blocks. One advantage of using a composite block is that resin can be less susceptible than ceramic to chipping during the milling process.2 Glass ceramic materials are stiff and brittle, making them highly susceptible to chipping and fracture.<sup>3</sup> Resin composite restorations do not cause excessive wear on natural dentition. In addition, if resin cement is used as the luting material, the modulus of elasticity of the cement is similar to that of the composite restoration, which can create a more uniform stress distribution throughout the teeth compared to ceramic inlays.<sup>4,5</sup>

The retention of an indirect restoration largely depends on the adhesive cementation. Composite resin cement with a multistep application technique has been used to bond restorations. The use of conventional resin cements can improve the physical properties of an indirect restoration; however, a multistep application can be technique sensitive. Recently, self-adhesive cements (SACs) that do not require pretreatment of the tooth surface have been introduced and are gaining popularity. Their major advantage is simple application with little technique sensitivity. SACs have a characteristic requirement of need for pH increase: when an SAC is mixed, it has an initially low pH to demineralize the tooth material. After a

certain amount of time, that initial acidity should be neutralized to produce a durable cement.

Another current trend in composite restoration is use of a one-step universal adhesive for dentin bonding. One-step self-etch universal adhesives make the bonding procedure simple and fast. However, they create several problems for composite restorations.<sup>8</sup> Moreover, incompatibility problems between one-step self-etch systems and self-/dualcure resin composites have been reported. 9,10 It is known that the tertiary amine used as the accelerator in self-/dual-cure composites can be neutralized by acidic functional monomers.9 Despite those incompatibility issues, the manufacturer of the nanoceramic block material recommends that its universal dentin adhesive be applied to both the restoration and the tooth before cementation. Therefore, it was questioned whether interfacial adaptation could be improved by applying a universal adhesive before placing the luting material. If the universal adhesive is to be applied, a flowable bulkfill composite could be used as the luting material. It was also wondered whether interfacial adaptation using the flowable bulk-fill composite differed from that cemented with SACs.

The clinical success of indirect restorations depends on a durable bond to the tooth material. <sup>11,12</sup> Several researchers investigated bond strength and fracture surfaces using light microscopes, scanning electron microscopes, or transmission electron microscopy. <sup>13-15</sup> In those studies, adhesive, mixed, or cohesive failure at different locations can imply a weak or strong interface in a cemented restoration. However, those studies were performed after interfaces had been detached or separated.

Optical coherence tomography (OCT) can be used to investigate the interfaces inside a restored tooth and to determine whether a microgap is present. When light passes through the interface between two types of media with different refractive indices, a portion of the light is reflected depending on the incidence angle and refractive index (n). The refractive index of air is 1.0 (n), that of water is 1.3, and that of a tooth or resin composite is 1.5-1.6. If there is a microgap between a tooth and a restoration, air or water can be present at the interface. When light transverses the air at the interface, a different portion of light is reflected, and the OCT system shows a higher signal intensity. Swept-source OCT (SS-OCT) is a specific type of OCT and is known to have high image resolution and scan speed. Therefore, SS-OCT can

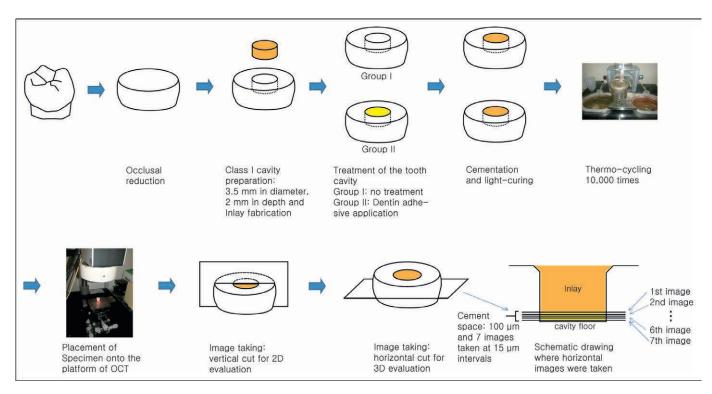


Figure 1. Experimental procedure for this study.

be used to investigate internal interfaces without damaging the specimen. 16

The first objective of this study was to determine whether there was a difference in interfacial adaptation when diverse luting materials were used for resin nanoceramic inlay cementation. The second objective was to investigate whether application of a universal dentin adhesive before placement of the luting material would improve the interfacial adaptation. The final objective of this study was to compare the two interfaces: inlay-side interface between the restorative material and the luting material and dentin-side interface between the tooth and the luting material.

The null hypotheses tested were as follows: 1) there is no difference in the interfacial adaptation of a resin nanoceramic inlay restoration when it is cemented by different luting materials; 2) when a resin nanoceramic inlay is cemented with self-adhesive cement after application of a universal dentin adhesive, the interfacial adaptation does not differ from that without pretreatment; and 3) when SAC is used as the luting material, there is no difference in interfacial adaptation between the restorative material and the luting material compared to the tooth and the luting material.

#### **METHODS AND MATERIALS**

#### **Specimen Preparation**

The setup used in this study is schematically illustrated in Figure 1. Extracted human third molars, free of cracks, caries, and restorations, were selected and stored in a 0.5% chloramine solution at 4°C and used within two months of extraction. Seventy-four teeth were chosen with a buccolingual dimension of  $10.5\pm0.5$  mm. The occlusal surface of each tooth was flattened with a trimmer and 320-grit SiC abrasive paper. Round class I cavities were prepared on the occlusal surface with a flat-end, straight diamond bur (6837-016, Komet Brasseler, Lemgo, Germany). The dimensions were  $3.5\pm0.2$  mm in diameter and  $2\pm0.1$  mm in depth. For cutting efficacy, a new bur was used every five preparations.

#### **CAD/CAM Inlay Fabrication**

Each prepared cavity was optically scanned (Medit Identica hybrid scanner, Medit, Seoul, Korea), and a virtual inlay was created with 100 μm of cement space (Milling software, Excad v 2.0.0.3). A resin nanoceramic block (21×19×14 mm) of Lava Ultimate (A2 HT, 3M ESPE, Neuss, Germany) was used to fabricate the inlays, which were milled using a

Roland Inlab milling machine (DWX-51D, Roland DG, Harmamatsu, Japan).

#### **Groups and Restorative Procedure**

For the control-1 group (PV5, n=6), the resin nanoceramic inlays were cemented using Panavia V5 (Kuraray Noritake, Tokyo, Japan). The internal surface of each indirect inlay was air-abraded using 50- $\mu$ m Al<sub>2</sub>O<sub>3</sub> particles 10 mm from the surface and two bars of (30 psi) pressure until the entire bonding surface had a matte appearance. After surface treatment, the particles were removed with alcohol, followed by ultrasonic cleaning in distilled water for three minutes and air drying. Then, Ceramic Primer Plus was applied and dried following the manufacturer's instructions. For tooth treatment, the Tooth Primer in the Panavia V5 kit was applied to the dentin surface of the tooth cavity. Following the manufacturer's instructions, gentle dry air was blasted for 20 seconds after Tooth Primer application. Equal amounts of the base and catalyst of Panavia V5 were mixed and placed into the tooth cavity. After positioning the fabricated inlay into the cavity, a load of 0.5 kg was applied on the inlay surface to allow the extrusion of excess cement. After a transparent celluloid strip was placed onto the top of the inlay, light-curing was performed (1200 mW/cm<sup>2</sup>, Elipar S10, 3M ESPE, St. Paul. MN. USA) on the top surface for 40 seconds.

For the control-2 group (FC2, n=6), Fuji Cem 2 (GC, Tokyo, Japan) was used for inlay cementation. After the same mechanical treatment on the inlay surface as used for the control-1 group, equal amounts of Fuji Cem 2 base and catalyst were mixed and placed into the tooth cavity. After positioning the inlay, the cement was allowed to set for 10 minutes without light-curing.

For the experimental groups, the teeth were randomly divided into two groups (groups I and II). Group I was composed of four subgroups, and the inlays were cemented without pretreatment. Group II was composed of six subgroups, and the inlays were cemented after application of a universal dentin adhesive. For group I, the fabricated inlay was cemented without pretreatment. The prepared teeth and fabricated inlays were randomly assigned to four subgroups (n=6 per subgroup) according to luting material. The cementing procedures are described in Table 1. For group RXU, RelyX U200 (3M ESPE) was used. For group GCL, G-Cem LinkAce (GC) was used. For groups SC2 and MLS, SmartCem2 (Dentsply Caulk, Milford, DE, USA) and Multilink Speed (Ivoclar Vivadent, Schaan, Liechtenstein) were used, respectively.

For group II, the fabricated inlay was cemented after application of a one-step self-etch universal adhesive, Clearfil Universal Bond Quick (CUB). The CUB was applied to the internal portion of the inlay and cavity dentin with a rubbing motion and dried by blowing mild air onto it until the adhesive did not move. The teeth and fabricated inlays in group II were randomly assigned to six subgroups (n=6 teeth per subgroup). For group RXU, RelyX U200 (3M ESPE) was used for cementation after CUB had been applied. For group GCL, G-Cem LinkAce (GC) was used after CUB treatment. For groups SC2 and MLS, SmartCem2 (Dentsply Caulk) and Multilink Speed (Ivoclar Vivadent), respectively, were used in the same way. For group SDR, the SDR (Smart Dentin Replacement, Dentsply Caulk) bulk fill flowable composite was used as the luting material after CUB treatment. For group VBF, Venus Bulk Fill flowable composite (Heraeus Kulzer, Dormagen, Germany) was applied in the same way.

After the luting material was applied, the fabricated inlay was placed. A load of 0.5 kg was applied on the occlusal surface to allow extrusion of excess cement. After a transparent celluloid strip was placed onto the top of the inlay, light-curing was performed (1200 mW/cm², Elipar S10, 3M ESPE) on the top surface for 40 seconds.

#### **Thermocycling Procedure**

All specimens were stored in water at room temperature for 24 hours before thermocycling. Then, the specimens were fatigued with 10,000 thermocycles to represent approximately one year of clinical functioning. The teeth were cycled between water baths of 5°C and 55°C with a dwell time of 30 seconds and a transfer time of five seconds. After thermocycling, the specimens were stored in water at room temperature.

#### SS-OCT System

The SS-OCT system (IVS-2000, Santec Co., Komaki, Japan) used in this study was a frequency-domain OCT system integrating a high-speed frequency. To sweep the external cavity, the near-infrared spectrum was swept from 1260 to 1360 nm, with its spectral bandwidth centered at 1310 nm, at a scan rate of 20 kHz. The system has a handheld probe with a power less than 5 mW. Backscattered light-carrying information about the microstructure of the sample was collected, digitized in a time scale, and analyzed in the Fourier domain to reveal information at each location on the x-axis or depth (A-scan). Combining a series of A-scans along a scan path can

Code	Material	Product	Manufacturer	Composition	Application Procedure
CUB	Self-etch universal dentin adhesive	Clearfil universal bond quick	Kuraray Noritake, Tokyo, Japan	Bis-GMA, 10-MDP, HEMA, hydrophilic amide monomer, filler, ethanol, water, NaF, photo initiators, chemical polymerization accelerator	After the dentin was dried, the adhesive was applied. Then, the entire cavity wall was dried by blowing mild air.
PV5	Dual-cure resin cement	Panavia V5	Kuraray Noritake, Tokyo, Japan	Paste A: Bis-GMA, TEGDMA, initiators, accelerators, silanated barium glass filler; Paste B: Bis-GMA, silanated aluminum oxide filler, camphorquinone	The Tooth Primer was applied, left for 20 seconds, and gently air-dried. The mixed paste was placed.
FC2	RMGI (resin modified glass ionomer) cement	Fuji Cem 2	GC, Tokyo, Japan	Paste A: Fluoroaluminosilicate glass, HEMA, dimethacrylate, pigment, initiator; Paste B: Polyacrylic acid, silica powder, initiator	Paste A and paste B were mixed for 10-15 seconds. The cavity wall was coated with the cement.
RXU	Self-adhesive cement	RelyX U200	3M ESPE, St Paul, MN, USA	Methacrylate monomers containing phosphoric acid groups, silanated fillers, initiator, alkaline fillers	Mixed cement was applied to the cavity using an auto-mix syringe.
GCL	Self-adhesive cement	G-Cem LinkAce	GC, Tokyo, Japan	Powder: Fluoroaluminosilicate glass, initiator, pigment; Liquid: 4- META, phosphoric acid ester monomer, UDMA	Mixed cement was applied to the cavity using an auto-mix syringe.
SC2	Self-adhesive cement	Smart-Cem2	Dentsply Caulk, Milford, DE, USA	UDMA, di- and tri-methacrylate resins, fluoroaluminosilicate glass, initiator, accelerators	Mixed cement was applied to the cavity using an auto-mix syringe.
MLS	Self-adhesive cement	Multilink Speed	Ivoclar Vivadent, Schaan, Liechtenstein	Methacrylate monomers containing phosphoric acid groups, dimethacrylate, barium glass, ytterbium trifluoride	Mixed cement was applied to the cavity using an auto-mix syringe.
SDR	Bulk-fill flowable composite	SDR	Dentsply Caulk, Milford, DE, USA	Modified UDMA, EBPADMA, TEGDMA, Ba-Al-F-B silicate glass, St-Al-F silicate glass, photo-initiator	Flowable composite was applied to the cavity
VBF	Bulk-fill flowable composite	Venus Bulk Fill	Heraeus Kulzer, Dormagen, Germany	UDMA, EBPADMA, Bis-EMA, Ba-Al-F silicate glass, YbF <sub>3</sub> , silicon dioxide	Flowable composite was applied to the cavity

Abbreviations: Bis-EMA, bisphenol-A polyethylene glycol diether dimethacrylate; Bis-GMA, bisphenol-A diglycidyl ether dimethacrylate; EBPADMA, ethoxylated bisphenol A dimethacrylate; HEMA, 2-hydroxyethyl methacrylate; TEGDMA, triethylene glycol dimethacrylate; UDMA, urethane dimethacrylate; 4-META, 4-methacryloyloxyethyl trimellitate anhydride; 10-MDP, 10-methacryloyloxydecyly dihydrogen phosphate.

produce a B-scan. By transforming the raw B-scan data into grayscale, a cross-sectional image can be created. Using serial B-scans over an area, the system produces a 3D image (horizontal cut image in this experiment). The axial resolution of the OCT system was 11  $\mu$ m in air, which was equivalent to 7  $\mu$ m in oral hard tissues and resin composites, assuming a refractive index of about n=1.5.

#### SS-OCT Image Collection and Analysis

Each specimen was positioned on the metal platform of the OCT system. The surface of the specimen was dried using an air duster to standardize the surface conditions. Using the handheld scanning probe connected to the SS-OCT, the light beam was projected onto the inlay surface of the specimen and scanned across the area.

For vertical-cut image collection, the first SS-OCT image was taken at the center of the restoration. The light beam was projected onto the bottom surface of the specimen and scanned across the area. The OCT probe was set at a fixed distance above the specimen. The first SS-OCT image of the restoration was taken parallel to the buccolingual plane of the cavity at the center of the restoration (Figure 1). After the first image was taken, the platform holding the specimen was moved 0.3 mm distally, followed by acquisition of the second image. The third image was taken after the platform was positioned at 0.3 mm mesially from the original center position. These vertical-cut

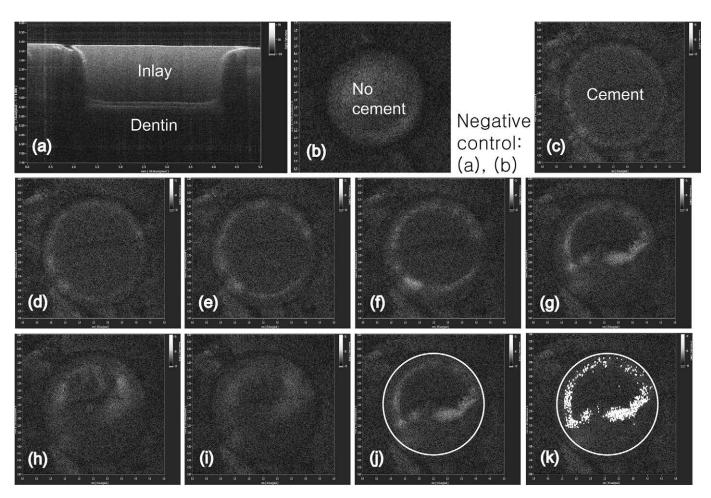


Figure 2. SS-OCT images for interfacial adaptation measurement. (a) Representative OCT image of a negative control without luting material: vertical cut. (b) A negative control image of the cement space: horizontal cut. (c) The first horizontal-cut image with luting material (PV5) in the cement space. The luting material shows an intensity value similar to that of dentin. The first image was taken parallel to the cavity floor 5 µm down from the inlay base. (d) The second image with PV5 in the cement space. The second image was taken 15 µm down from the first image. (e-i) The third, fourth, fifth, sixth, and seventh images, respectively, which were each taken 15 µm down from the previous image. j and k are the same images as in g, which are presented for the calculation of HB% of image g. (j) The same image as in g with a circle representing the cavity border. (k) The same image as in j processed by GapAnalyzer. The white dots on image k are brighter pixels that have higher signal intensity than the threshold to indicate a microgap at the cement space. The signal intensity of the threshold value was determined using the negative control image. On image k, the area of white dots was measured and then divided by the area of the circle to calculate the interfacial adaptation (HB%). On image k, the HB% was calculated to be 11.9%.

images were used as references and not applied in the statistical analysis.

For horizontal-cut image collection, the first SS-OCT image of the restoration was taken parallel to the cavity floor 5  $\mu m$  below the inlay base. After the first image was taken, the platform holding the specimen was moved upward 15  $\mu m$ , followed by acquisition of the second image. A total of seven images were taken in the cement space of each specimen at 15- $\mu m$  intervals (Figure 1).

To evaluate the resin-tooth interfacial adaptation, raw OCT data were imported into image analysis software (ImageJ, ver. 1.48, National Institutes of Health, Bethesda, MD, USA). If air or water was present at the tooth-restoration interface, part of the

light reflected from the interface was visualized as a bright spot on the image (Figure 2). The high brightness (HB%) parameter was defined as the percentage of brighter pixels with a signal intensity above the threshold value in the signal intensity profile and calculated to indicate microgap or nonadapted area in the cement space (Figure 2k). To calculate the percentage of pixels brighter than the threshold, images were processed using GapAnalyzer, plug-in software that uses a binarization process described previously. <sup>19,20</sup> Using the seven horizontal-cut cross-sectional images of each specimen, the mean HB% per sample was calculated. A higher HB% represents inferior interfacial adaptation in the cement space.

Group Control <sup>b</sup>			Experimental Subgroup						
	PV5	FC2	RXU	GCL	SM2	MLS	$\begin{array}{c}Adhesive^{\pmb{c}}\\ + SDR\end{array}$	$\begin{array}{c} Adhesive^{\pmb{c}} \\ + VBF \end{array}$	
I: Cementation without pretreatment	10.2 (3.4) a	24.6 (4.6) d	10.7 (3.4) a,b,A	11.1 (3.6) a,b,A	18.2 (4.0) c,B	13.0 (3.8) a,b,B			
II: Cementation with dentin adhesive	10.2 (3.4) a		9.4 (3.2) a,A	10.4 (2.9) a,A	14.8 (3.4) c,A	10.4 (3.0) a,A	11.1 (3.2) a,b	12.9 (3.5) b,c	

<sup>&</sup>lt;sup>a</sup> Standard deviations are indicated in parentheses. In each row, values marked by identical lowercase letters are not significantly different (one-way ANOVA and Tukey test, p>0.05). In each column, values marked by identical uppercase letters are not significantly different (independent t-test, p>0.05).

### Comparative Analysis of the Inlay-side Interface and Dentin-side Interface

The CAD/CAM inlay was fabricated with a 100-um cement space. For inlay-side interfacial adaptation, which can represent the interface between the inlay and luting material, the first and second horizontalcut images were measured (Figure 1). As explained earlier, the first image was taken parallel to the cavity floor 5 µm down from the inlay base (Figure 2c). The second image was taken 15 µm down from the previous image (Figure 2d). The inlay-side interfacial adaptation was the average HB% of the first and second images. The dentin-side interfacial adaptation, which may represent the interface between the tooth and the luting material, was calculated as the mean of the sixth and seventh images. Inlay-side and dentin-side interfacial adaptations were calculated for each specimen.

#### **Statistical Analysis**

Because the distribution of measurements in each group was normal (Shapiro-Wilk test, p>0.05), parametric statistical tests were performed. To test for homogeneity of variances of the groups, Levenes test was used (p>0.05). The HB% of the SAC groups

(RXU, GCL, SM2, and MLS) was analyzed using two-way analysis of variance (ANOVA) to test the effects of universal adhesive application and luting material, as well as their interaction. For the comparison of subgroups in groups I and II, one-way ANOVA and the Tukey test were performed. To compare the SAC subgroups between groups I and II, an independent t-test was used (Table 2). For analysis of interfacial adaptation at the inlay-side and dentin-side interfaces, one-way ANOVA and post hoc Tukey tests were performed (Table 3). All statistical analyses were conducted using PASW Statistics 18 software (SPSS for Windows, SPSS, Inc, Chicago, IL, USA) with the significance level set at  $\alpha$ =0.05.

#### **RESULTS**

The interfacial adaptations (HB%) are summarized in Table 2. The two-way ANOVA showed the significance of universal adhesive application, the luting material, and their interaction, at 0.008, less than 0.001, and 0.036 (p<0.05), respectively. After one-way ANOVA and Tukey analysis, interfacial adaptation was different depending on the luting material in both groups I and II (Table 2; each row,

Group	Control, PV5 <sup>b</sup>	Experimental Subgroup					
		RXU	GCL	SC2	MLS		
Inlay-side interface	9.1 (1.6) a,A	9.8 (4.0) a,A	11.7 (4.6) a,A	17.2 (2.0) b,A,B	13.0 (2.4) a,B		
Dentin-side interface	11.1 (2.3) a,A	10.6 (2.2) a,A	11.1 (2.5) a,A	19.1 (3.6) b,B	12.6 (4.4) a,A,E		
II							
Inlay-side interface	9.1 (1.6) a,A	11.4 (3.8) a,A	11.0 (2.5) a,A	15.9 (3.0) b,A,B	11.3 (3.3) a,A,E		
Dentin-side interface	11.1 (2.3) a,b,A	8.3 (2.0) a,A	8.8 (2.1) a,A	14.1 (3.6) b,A	9.1 (2.4) a,A		

<sup>&</sup>lt;sup>a</sup> Standard deviations are indicated in parentheses. In each row, values marked by identical lowercase letters are not significantly different (one-way ANOVA, p>0.05). In each column, values marked by identical uppercase letters are not significantly different (one-way ANOVA, p>0.05).

b The results of PV5 (control-1) for Groups I and II and FC2 (control-2) for Group I are presented for comparison with other luting materials.

<sup>&</sup>lt;sup>c</sup> The adhesive was CUB.

The results of PV5 in Groups I and II are presented for comparison with other subgroups.

p<0.05). The independent t-test showed that application of a universal dentin adhesive significantly improved adaptation in some of the SAC groups (Table 2; each column, p<0.05).

In the comparison of inlay-side and dentin-side interfaces in groups I and II, no different interfacial adaptation was found (Table 3; p>0.05). In the comparison of inlay-side interfacial adaptations between groups I and II, no difference was found (Table 3; first and third rows, p>0.05). However, a different adaptation was found at the dentin-side interface between a subgroup of group I and a subgroup of group II (Table 3; second and fourth rows, p<0.05).

#### DISCUSSION

The first null hypothesis was rejected because HB% (internal adaptation) was different depending on luting material. Some of the SACs showed interfacial adaptation similar to that of the control group, whereas other SACs had inferior interfacial adaptation. Thus, SACs' self-adhesive capacities can be different by product features. 21 SACs have a complex composition, including methacrylate monomers, acidic functional monomers, fillers, and initiators (Table 1). The selection and proportion of each component may determine the final properties of the materials.<sup>22</sup> The second null hypothesis was partially rejected because application of a universal dentin adhesive improved interfacial adaptation, but only in some subgroups. The third null hypothesis was accepted because the inlay-side and dentin-side interfaces showed no difference in interfacial adaptation.

SACs are reported to have a limited capacity to demineralize dentin and have no significant infiltration more than a micrometer into the dentin.<sup>23</sup> However, some SACs showed interfacial adaptation comparable to that of conventional resin cement (Table 2). It was previously reported that RXU can make cement bind with the calcium in hydroxyapatite due to the phosphate group in its functional monomer.<sup>24</sup> GCL is known to contain two functional monomers: 4-META (4-methacryloyloxyethyl trimellitate anhydride) and a phosphoric-acid ester monomer. 25 4-META can be hydrolyzed to form 4-MET (4methacryloxyethyl trimellitic acid), which can have a chelating reaction with hydroxyapatite. 26,27 SC2 and MLS had relatively inferior adaptation (Table 2). In other studies, SC2 and MLS showed relatively low micro-tensile or shear bond strength to dentin and high pretest failures. 22,28,29 One study indicated that the bonding ability of SACs depends mainly on the presence of an acid-functionalized monomer.<sup>30</sup> Thus, SACs' adhesive properties are material dependent and could be related to the composition of each material.<sup>21,28</sup>

SAC manufacturers have challenges to address. SACs need two opposite characteristics to achieve stable cementation: hydrophilicity for tooth bonding and hydrophobicity for resin matrix durability. For the bonding mechanism, SACs include acidic, hydrophilic monomers that can chemically interact with the hydroxyapatite in the tooth. Carboxylic or phosphoric acid groups in the monomers, which can show a low pH before reaction, form a complex with the calcium of hydroxyapatite. 23,31 To neutralize this initially acidic condition, a glass-ionomer concept was adopted.<sup>32</sup> The acid-base reaction of the acidic functional group with the basic inorganic filler in the cement or the mineralized tooth surface can lead to neutralization.<sup>30</sup> However, some SACs still have low pH values (less than 4) even two to seven days after mixing. 31,33 This acidity could compromise the curing of the resin, increase water sorption, and lower the SAC's mechanical properties. 30,34 Another problem for SAC bonding can be dependence on the condition of the bonding substrate.35 SACs showed different bond durability depending on dentin surface conditions such as moisture and smear layer.<sup>29</sup>

Despite the problems with one-step universal adhesives, application of the universal adhesive produced similar or better interfacial adaptation than not using it (Table 2). Applying a universal adhesive to dentin provides an acidic treatment to the dentin. In terms of pH, SACs could be applied to the adhesive-treated surface because the acidic functional monomer in the SAC has a mechanism similar to that of a self-etch universal dentin adhesive.<sup>30</sup> When using a universal dentin adhesive in this study, one reason for the improved adaptation could be modification of the smear layer on the dentin surface. The presence of a smear layer has been recognized as a weak link to dentin.<sup>36</sup> It was reported that mild acidic treatment, which could remove the superficial, loosely bound fraction of the smear layer, could enhance adhesion.<sup>23</sup> Some SACs may not be able to modify the smear layer, so an additional treatment of universal adhesive could improve interfacial adaptation.

No differences in adaptation were found in the comparison of the inlay-side and dentin-side interfaces in groups I and II (Table 3). Generally, the dentin-side interface showed inferior adaptation to the inlay-side; however, no statistically significant difference was found. For the comparison of the

inlay-side of group I and that of group II, no difference was found. It could indicate that universal adhesive treatment on the resin inlay surface did not significantly improve adaptation at the inlay-side interface. It was previously reported that mechanical treatments such as sandblasting would be the most determining factor for improving the retention of indirect composite restorations. 22 Another paper presented that sandblasting Lava Ultimate contributed most of the bonding, whereas the effects of other treatments remained unclear.<sup>13</sup> In the comparison of dentin-side interfaces between groups I and II, one significant difference was found (Table 3). It can suggest that a universal adhesive may improve adhesion to the dentin interface depending on the luting material.

The horizontal OCT images show the interfacial debonding pattern in the cement space under the inlay. The most frequent debonding pattern at the inlay side was circular along the border of the cavity, while that at the dentin side was irregular (Figures 3 and 4). The circular pattern can be due to polymerization shrinkage of the luting material toward the center of the cement space and the relatively even bond strength on the inlay-side interface. However, the debonding pattern on the dentin side was somewhat different and frequently irregular, which might indicate that bonding to the dentin surface was not consistent. Several other variables might also affect interfacial adaptation at the dentin surface: different characteristics of regional dentin, different properties of dentin adhesives, surface defects, air bubbles, phase separation, and a nonuniform adhesive layer. Those variables could make the bond strength on the dentin side nonuniform to produce an irregular debonding pattern.

Drawbacks and limitations should be considered when the interfacial adaptation is evaluated by the SS-OCT image. First, SS-OCT has depth limitations. Even though SS-OCT shows very clear images within the penetrating depth of the laser, it cannot be used in deep cavities or with a non-lighttransmitting material such as metal. The imaging depth of SS-OCT systems had been reported to be in the range of 2-3 mm.<sup>37</sup> Therefore, the cavity design for SS-OCT evaluation can be limited depending on the light penetration capability. The second thing to consider is the threshold for the gap decision. It would be ideal to evaluate the images with one signal intensity threshold. However, defining a SS-OCT threshold was very complicated because the signal intensity was affected by the light intensity, scattering, attenuation, and transmittance properties of the material. 16,19 In this experiment, the threshold was determined after the signal intensity was measured on the negative control image.

PV5, which is a dual-cure resin cement, was used as a control. It is claimed that PV5 contains a novel amine-free redox initiator that makes it possible to avoid the oxidation of amine. Unlike the previous version (Panavia F2), PV5 does not contain 10-MDP in the cement paste, but it is included in the Tooth Primer. Essentially, 10-MDP is a hydrophilic, acidic monomer that can lead the cement to a low degree of conversion. FC2, which was used as a second reference control, showed much more debonding on the dentin side than on the inlay side. For the SDR and VBF groups, a bulk-fill flowable composite which has no acid-base reaction for polymerization, was tested as the luting material. Those groups showed interfacial adaptation comparable to those of the resin-based cements (Table 2). However, the debonding pattern was somewhat different and frequently showed an agglomerated pattern. Bulkfill flowable composites do not have self-curing potential and are fully light dependent, highlighting the importance of transmitting light through the whole restorative material.

SACs are known to contain specific components to prevent the acid-base reaction problem.<sup>30</sup> As explained in the Introduction, the problem with self-/ dual-cure resins is incompatibility between the residual acidic monomer of the universal dentin adhesive and the tertiary amine catalyst.38 For SACs, an acid-tolerant oxidant such as cumene hydroperoxide is included in the acidic part, and a reductant such as benzoyl thiourea is used in the nonacidic part. 30,39 Adverse acid-base reactions can also be circumvented by using acid-resistant catalysts such as aryl sulphate salts. 9,30,40 The sodium salt of aryl sulfinic acid can react with acidic monomers to produce phenyl free radicals, which can initiate the self-curing process of the resin.<sup>40</sup> This acid-tolerant composition can help to lessen the adverse acidic effects of the universal adhesive.

Immediate light-curing can be important to reduce adverse effects when using a one-step self-etch universal adhesive. Studies have shown that light-curing improved the degree of conversion and bond strength compared with self-curing alone. 41-43 Photo-initiators such as camphorquinone (CQ) and diphenyl-(2,4,6-trimethylbenzoyl) phosphine oxide (TPO) are used in the acidic components of SACs. After acid contamination, light-curing of resin was inhibited to a much lower extent than that of self-curing, possibly because photo-initiation of free radicals is

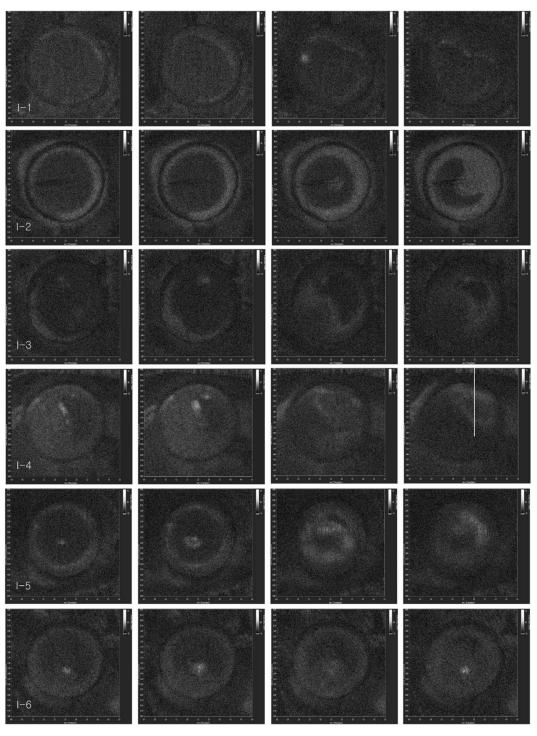


Figure 3. Interfacial horizontal-cut images of group I at the cement space. Row I-1 represents the first image, second image, sixth image, and seventh image of the control-1 group (PV5). Row I-2 represents the same four images of control-2 group (FC2). Row I-3 represents the same four images from the RelyX U200 group (RXU). Row I-4 represents the same four images from the G-cem LinkAce group (GCL). Row I-5 represents the same four images from the Multilink Speed group (MLS).

much faster than that of chemical initiators.<sup>44</sup> Another problem of one-step adhesives is that the permeable adhesive layer can trigger osmotic fluid movement.<sup>8</sup> When the adhesive layer was not

immediately cured, water sorption from the dentinal tubule increased. <sup>44-46</sup> It is thought that immediate light-curing with adequate light should be performed whenever a universal dentin adhesive is applied.

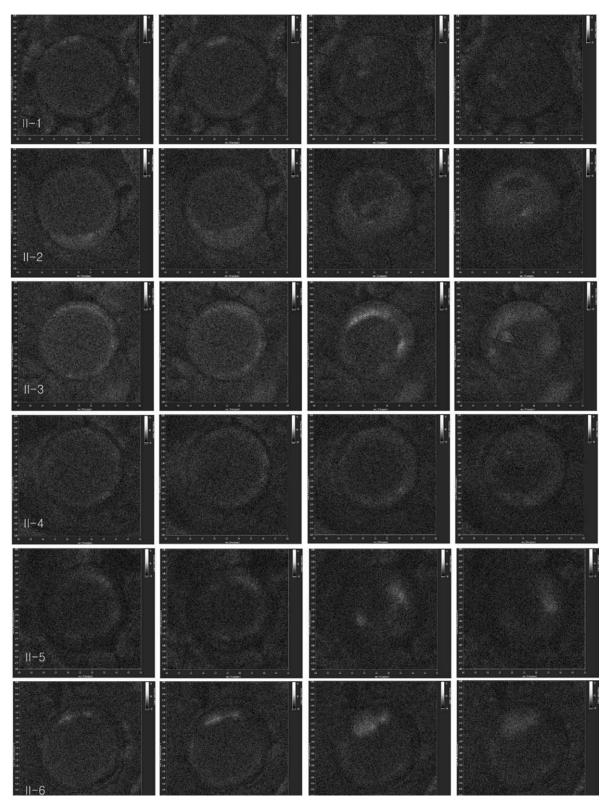


Figure 4. Interfacial horizontal-cut images of group II at the cement space. Row II-1 represents the first image, second image, sixth image, and seventh image of the RelyX U200 group (RXU). Row II-2 represents the same four images from the G-cem LinkAce group (GCL). Row II-3 represents the same four images from the Smartcem2 group (SC2). Row II-4 represents the same four images from the Multilink Speed group (MLS). Row II-5 represents the same four images when the inlays were cemented by SDR flowable resin after application of the universal dentin adhesive. Row II-6 represents the same four images when the inlays were cemented by VBF flowable resin after application of the universal dentin adhesive.

#### CONCLUSION

Under the limitations of this study, it can be concluded that the interfacial adaptation of a resin nanoceramic inlay restorations were different depending on luting material. For the resin nanoceramic inlay cementation, the application of a universal dentin adhesive before SAC placement showed similar or better interfacial adaptation than without the dentin adhesive. When SAC was used as a luting material without any pretreatment, the interfacial adaptation of the inlay-side was not different from that of the dentin-side interface. The comparison of the interfacial adaptation with the adhesive and that without the adhesive was performed on the inlay-side and the dentin-side. On the inlay-side, the interfacial adaptation with the universal adhesive application was not different from that without the adhesive. However, on the dentinside, the interfacial adaptation with the adhesive was better in some groups.

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#### **Regulatory Statement**

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of St Vincent Hospital, the Catholic University of Korea. The approval code issued for this study is VC17OESI0173.

#### **Conflict of Interest**

The authors of this manuscript certify that they have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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Associate Professor/Professor and Head Department of Restorative Dentistry University of Illinois at Chicago College of Dentistry

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#### Time-dependent Microhardness Gradients of Self-adhesive Resin Cements Under Dual- and Selfcuring Modes

T Geng • Y Pan • ZZ Liu • C Yuan • P Wang • X Meng

Clinical Relevance: Acid-functional monomers in self-adhesive resin cements may decrease their self-curing polymerization ability. Light irradiation optimizes polymerization performance.

https://doi.org/10.2341/19-006-L

## Effectiveness of Whitening Strips Use Compared With Supervised Dental Bleaching: A Systematic Review and Meta-analysis

GRV da Rosa • BM Maran • VL Schmitt • AD Loguercio • A Reis • FS Naufel

Clinical Relevance: Bleaching performed at home while under the supervision of a dentist provides greater color alteration compared with whitening strips when evaluated with a spectrophotometer, although the color alteration was undetectable by unaided human eyes.

https://doi.org/10.2341/19-160-L

## Polymerization Stress and Gap Formation of Self-adhesive, Bulk-fill and Flowable Composite

EL Nakano • ASC de Souza • LCC Boaro • LH Catalani • RR Braga • F Gonçalves

Clinical Relevance: Bulk-fill materials show a similar or better performance than control flowable materials regarding interfacial integrity. However, some self-adhesive composites need improvements to achieve competitive performance.

https://doi.org/10.2341/19-166-L

#### **Clinical Efficacy of Different Dentin Desensitizers**

GB Eyüboglu • P Naiboglu

Clinical Relevance: Teethmate Desensitizer, Clinpro White Varnish, Shield Force Plus, and Gluma could be recommended for treating dentin hypersensitivity in terms of clinical efficacy.

https://doi.org/10.2341/19-258-C

#### **Effect of Tribochemical Coating on Composite Repair Strength**

AV Ritter • T Sulaiman • A Altitinchi • F Baratto-Filho • CC Gonzaga • GM Correr

Clinical Relevance: Tribochemical treatment of existing composite surfaces is highly effective for composite repair. When repairing an old composite restoration, the clinician should try to use the same composite originally used for the restoration. If the information about the original restoration is not known, a composite with strong mechanical properties should be used for the repair restoration.

https://doi.org/10.2341/19-145-L