Microleakage of “Bulk-Fill” Composite Resin for Class II Restorations Pretreated With CO2 Laser in Deciduous Molars: An In Vitro Study

Larissa Costa Santos¹, Ravana Angelini Sfalcin², Eugenio José Garcia³, Fátima Antônia Aparecida Zanin¹, Aldo Brugnera Junior⁴, Daniela Fátima Teixeira Silva³, Anna Carolina Ratto Tempestini Horlina⁴, Kristianne Porta Santos Fernandes², Raquel Agnelli Mesquita-Ferrari², Sandra Kalil Bussadori²,³*

¹Department of Biophotonics Applied to Health Sciences, University Nove de Julho (UNINOVE), Vergueiro St, 235, Liberdade, São Paulo, Brazil
²Postgraduate Program in Biophotonics Applied to Health Sciences, University Nove de Julho (UNINOVE), Vergueiro St, 235, Liberdade, São Paulo, Brazil
³Postgraduate Program in Program in Rehabilitation Sciences, University Nove de Julho (UNINOVE), Vergueiro St, 235, Liberdade, São Paulo, Brazil
⁴Postgraduate Program in Laser in Dentistry, Federal University of Bahia (UFBA) and Founder of Brugnera and Zanin Institute, São Paulo, Brazil
⁵Biomedicine Engineering Division, Camilo Castelo Branco University, Estrada Dr altino Bondesan, 500, Eugênio de Melo, São José dos Campos, Brazil

Abstract

Introduction: Microleakage has been reported to cause dentin hypersensitivity because of the passage of bacteria and their products through the restoration-tooth interface and is one of the main reasons for replacement of restorations. CO2 laser can be used for treatment of dentin hypersensitivity. Thus, this study aimed to evaluate in vitro the microleakage in composite restorations following surface pretreatment with acid etching and CO2 laser.

Methods: Twelve human caries-free primary molars were selected. Class II cavities were prepared on occlusal mesial and occlusal distal surfaces. Specimens were randomly divided into four groups (n = 6): Group 1 (G1) – 37% phosphoric acid gel etching + Beautiful-Bulk Restorative – Giomer (Shofu Inc); Group 2 (G2) – 37% phosphoric acid gel etching + SDR Bulk-Fill Flow (Dentsply); Group 3 (G3) – CO2 laser irradiation + Beautiful-Bulk Restorative – Giomer (Shofu Inc); Group 4 (G4) – CO2 laser irradiation + SDR Bulk-Fill Flow (Dentsply). Surfaces were restored with bonding agent (Natural Bond DE, DFL). Specimens were cut longitudinally and immersed in 0.5% methylene blue solution for 4 hours. Microleakage scores were assessed under a magnifying glass at x3.5 and qualitatively analyzed by scanning electron microscope (SEM). Data were analyzed using nonparametric Wilcoxon test (P < 0.05).

Results: Scores prevailed between 0 and 2, however, no statistically significant difference was found among the groups (P = 0.05).

Conclusion: It could be concluded that all composite resins bulk fill did not show significant difference among them regarding microleakage using either CO2 laser or 37% phosphoric acid etching.

Keywords: Acid etching; Primary tooth; Composite resins, CO2 laser.

Introduction

Clinical success of composite resin restorations on posterior teeth depends on the quality of the bond and the adaptation of the material to the walls of the cavity. Methacrylate-based composites are routinely used in direct restorative procedures. Composites are considered sensitive materials that need to be inserted in increments of up to 2 mm to allow sufficient light penetration for polymerization, resulting in enhanced physical and mechanical properties. The main harmful effect of this conversion process is shrinkage of the material due to polymerization. Shrinkage is manifested as stress at the bond with the wall of the cavity, which can lead to interfacial defects, enamel fractures, cuspal movements and micro-cracks. Thus, the control of shrinkage stress in dental composites is essential to ensure margin integrity and the longevity of the restoration. The incremental technique minimizes
harmful polymerization shrinkage due to the lower C-factor during the polymerization of each layer.\textsuperscript{6,8} However, the incremental method has disadvantages, such as the possibility of incorporating voids or contamination between composite layers, resulting in possible bond failures between increments. Moreover, the time required to place and polymerize each layer is longer in comparison to a bulk-filling technique.\textsuperscript{9-11}

Recently, a new category of composites with modified chemical compositions to reduce polymerization shrinkage has been marketed for bulk application in direct posterior composite restorations. Bulk-fill composites can be inserted in a single layer measuring 4 to 5 mm in thickness and cured in a single step with a lower polymerization shrinkage and consequent stress.\textsuperscript{12,13} This allows a significant reduction in the time required to perform a restoration,\textsuperscript{14} which is very interesting to the pediatric dentistry attendance. Studies have evaluated the performance of bulk-fill composites in cavities such as Class II MOD preparations\textsuperscript{15} and authors have found a similar or even better performance of bulk-fill restorations comparing to traditional composite resins placed incrementally.\textsuperscript{15-17}

Giomers have recently been introduced, which are resin restorative materials that maintain the clinical advantages of glass ionomer cements. It has properties of both glass ionomer (fluoride release and fluoride recharge) and resin composites (excellent esthetics). The development of pre-reacted glass-ionomer filler that can be incorporated into resinous materials compensates its poor esthetics and dehydration potential.\textsuperscript{18} Surface pre-reacted glass ionomer particles (S-PRG) are currently incorporated in bulk-fill technologies, such as a low viscosity bulk-fill flowable giomer material (Beautiful Bulk Flowable, Shofu Dental Corporation, San Marcos, CA, USA) and a high viscosity bulk-fill giomer resin restorative material (Beautiful Bulk Restorative, Shofu Dental Corporation). Besides the polymerization shrinkage, dentin hypersensitivity is one of the major challenges in dental practice.\textsuperscript{19} The hydrodynamic theory\textsuperscript{20,21} explains the phenomenon of dentin hypersensitivity as an increase in the flow of the fluids present in dentinal tubules that have patent orifices, thereby activating nerves situated in the outer layers of the pulp. Microleakage may occur in the gap between the restorative material and tooth structure, and may cause dentin hypersensitivity. The techniques used to evaluate this phenomenon demonstrate that microleakage is not uniform along the interface due to different factors related to the quality of restorations, such as smear layer, acid etching, moisture and polymerization of the resin.\textsuperscript{22}

The use of lasers in dentistry has increased in recent years. Studies have shown effects such as the merging of intertubular dentin, the obliteration/opening of dentinal tubules, the re-crystallization of dentin and removal of the smear layer.\textsuperscript{23,24} On enamel, laser produces micro-explosions during hard tissue ablation that result in microscopic and macroscopic irregularities, making the enamel surface micro-retentive, thereby offering an adhesion mechanism without the need for acid etching.\textsuperscript{25} Pashley et al\textsuperscript{26} concluded that on dentin, laser treatment of smear layers will vaporize organic constituents but fuse mineral components together, thereby increasing the cohesive strength of the smear layer, and that those systems that etch dentin attempt to increase ability of hydrophilic resins to penetrate or permeate into dentin, and lasing conditions that decrease dentin permeability may lower the bond strength of such systems while increasing the bond strength of systems that bond to relatively impermeably smear layers.

The combination of carbon dioxide (CO\textsubscript{2}) laser with bulk-fill technologies could improve adhesion and minimize the frequency of gaps and microleakage. However, this aspect has not yet been fully explored. Thus, the present study aimed to evaluate, \textit{in vitro}, the microleakage after using low-shrinkage resins (low viscosity and high viscosity bulk-fill composites) for Class II restorations previously treated with either acid etching or CO\textsubscript{2} laser.

Materials and Methods

Twelve human primary molars, sound, caries free were selected. Teeth were clinically extracted at the UNIMES clinic-school and stored in distilled water to prevent dehydration. Twenty-four class II cavities were performed on mesial and distal faces (4 mm depth, 3 mm buccal/lingual or palatal length, 2 mm mesioaxial width) showing gingival cavosurface margins. Cavities were created conventionally on the occlusal/mesial and occlusal/distal surfaces (vertical slot) with a cylindrical diamond burr #1091 (KG Sorrensen) on high-speed dental drill. Samples were randomly divided into 4 groups (n = 6) as follows:

- **Group 1 (G1):** 37% phosphoric acid gel etching (Super Etch – SDI) for 15 seconds, rinsing, air drying, application of adhesive (Natural Bond DE, DFL) and restoration with Beautifil-Bulk Restorative – Giomer (Shofu Inc);
- **Group 2 (G2):** 37% phosphoric acid gel etching (Super Etch – SDI) for 15 seconds, rinsing, air drying, application of adhesive (Natural Bond DE, DFL) and restoration with SDR Bulk-Fill Flow (Dentsply);
- **Group 3 (G3):** CO\textsubscript{2} laser irradiation etching, application of adhesive (Natural Bond DE, DFL) and restoration with Beautifil-Bulk Restorative – Giomer (Shofu Inc);
- **Group 4 (G4):** CO\textsubscript{2} laser irradiation etching, application of adhesive (Natural Bond DE, DFL) and restoration with SDR Bulk-Fill Flow (Dentsply).

Application of adhesive and restoration were carried out according to the manufacturer’s instructions. Irradiation on the internal walls and gingival cavosurface margins was performed using a CO\textsubscript{2} laser (Ultralase 30, South Inc., USA), ultra-pulse module, focused beam, as
shows Table 1.

The handpiece was aligned perpendicular to cavity surface and moved by hand continuously over the area at approximately 2 mm/s during the exposure period to simulate a clinical laser etching technique.

The specimens were sealed with a layer of nail varnish applied over the entire surface, leaving an exposure window beyond 2 mm from margin of the restoration. Specimens were then immersed in 0.5% methylene blue dye, pH 7.2, during four hours under darkness, followed by rinsing in running water for 10 minutes.

Teeth were then sectioned longitudinally in a distal-mesial direction through the center of the restoration using a modified, water cooled, low-speed diamond saw (American Burrs, RS, Brazil), thereby obtaining 48 specimens. Specimens were observed with a magnifying glass of 3,5x by 2 calibrated examiners. The examiners analyzed twelve sections from each group using a numeric scale to determine the degree of marginal leakage based on the penetration of methylene blue dye at the tooth-restoration interface.

Spearman test was used to determine inter-examiner agreement. Microleakage data were submitted to the non-parametric Wilcoxon test ($P=0.05$). Scanning electron microscopes (SEM) were qualitatively analyzed.

Results

Microleakage Evaluation

Microleakage scores prevailed between 0 and 2, but ranged from 0 to 4 (Figures 1 and 2). Statistical nonparametric test Kruskal-Wallis was used to compare groups considering the results obtained by each examiner separately ($P=0.05$). Although scores have ranged from 0 to 4, statistical analysis did not show difference among the groups due to the distribution of the specimens, which showed a prevalence of microleakage scores between 0 to 2 (Figures 1 and 2).

To compare the groups between the examiners, Wilcoxon test was performed. According to the results, there was no statistical difference among groups ($P=0.05$) (Figure 3).

Scanning Electron Microscopy

Groups which showed higher variation were conventional composite resins bulk fill groups (Giomer), as shown by qualitative analysis using the microographies obtained by SEM (Hitachi TM3000 Tabletop, Tokyo, Japan), with 50x magnification at 7.5 kV. Figures 4A and 4B show scores 1 and 3, respectively, obtained from the specimens of G1 and G3. However, to the composite resin bulk fill flow groups (G2 and G4), micrographies showed higher adaptation of the material in Class II cavities, which presented a higher prevalence of score 0 than when conventional composite resin bulk fill was used (Figures 5).

Discussion

In the present study, microleakage was investigated...
using two different types of bulk-fill composite resins (conventional and flow) and 2 types of surface treatment (37% phosphoric acid and CO_2 laser etching).

Microleakage is an important property used to assess the success of restorative materials, as it demonstrates the possibility of the chemically undetectable passage of bacteria, molecules, fluids or ions between a restorative material and the walls of a cavity. Tracer dyes are available for microleakage studies and the difference in penetration among fuchsin, silver nitrate and methylene blue seems not to be significant. Methylene blue is one of the most common tracers and can be used at different concentrations. In the present study, methylene blue was used with the following scored evaluation criteria: 0) no microleakage; 1) microleakage only in enamel or less than 1/3 of the gingival wall in dentin; 2) up to the dentinoenamel junction or 2/3 of gingival wall in dentin; 3) reaching gingival wall in enamel and dentin; and 4) reaching the axial wall.

Class II cavities have been studied by several authors and this type of cavity has a gingival margin in dentin and enamel, which is a determinant factor for the occurrence of infiltration by marginal leakage. Problems commonly associated with shrinkage generated by the polymerization process and the cross-linking of monomers include infiltration of the restoration margins, secondary caries, enamel cracks and postoperative dentin hypersensitivity. To minimize such problems, restorative materials have appeared on the market with physical and mechanical properties designed to dissipate stress better, thereby causing less leakage. Restorative materials with low shrinkage stress are denominated ‘bulk-fill’ composites, which are able to fill the cavity in a single layer and with greater ease, making the procedure much faster, simpler and more practical. Moreover, the formulation of these materials allows for modulation of the polymerization reaction by use of special, stress-relieving monomers, the use of more reactive photoinitiators and the incorporation of different types of fillers, such as pre-polymer particles and fiberglass rod segments. Furthermore, bulk placement prevents void incorporation and contamination between composite layers, leading to more compact fillings.

Microleakage occurred in all groups in the present study. Although there was no statistically significant difference among the groups, specimens that received SDR flow had slightly lower scores than those that received the Giomer, independently of whether the cavity was prepared with 37% phosphoric acid or CO_2 laser etching. A previous study found that flowable resins, particularly low-shrinkage flowable composites (Surefil SDR flow), lead to significantly better results regarding microleakage at dentinal margins. On the other hand, the authors found no significant difference between a nanohybrid composite and low-shrinkage flowable composites at enamel margins, which confirms that the quality of adhesion to enamel is able to overcome curing shrinkage regardless of the volumetric shrinkage of the resinous material employed. The present findings corroborates with such data, as lower scores were found when using SDR Flow in comparison to the conventional bulk-fill Giomer. This can be explained by lower stress due to the low elastic modulus and lower wettability. In another study, Moorthy et al. rated the degree of marginal leakage with the use of or non-use of low shrinkage flow resin (bulk-fill flowable) in class II premolar cavities and found no significant difference in cervical microleakage, which also corroborates with the present findings. Besides, the fluid filling resins have a lower concentration of charge, great flow with excellent adaptation to the cavity with excellent adaptation to the cavity and low modulus of elasticity, which would support and better dissipate stress generated by thermal and masticatory stresses and favouring marginal sealing.

The use of laser irradiation is becoming widespread in dentistry. CO_2 laser has been employed in laboratory and clinical trials studies for hard and soft dental tissues. On hard dental tissues, CO_2 laser induces physical, chemical and morphological changes, such as the melting of enamel prisms and re-crystallization, with the formation of pores and small bubble-like inclusions. Previous clinical trials...
involving laser etching have shown promising results and authors have demonstrated that CO₂ laser causes no damage to the pulp of human teeth when less than 4 J of energy is administered to the enamel surface.

In our study, CO₂ laser irradiation was performed on enamel and dentin surfaces with 3 W and a pulse duration of 5000 µs. The results demonstrated no significant difference between CO₂ laser and 37% phosphoric acid etching with regard to subsequent microleakage following restorations. These findings corroborate with data described in previous studies, in which bond strength following laser etching proved to be comparable to that following acid etching, as demonstrated by the absence of statistically significant differences in microleakage for all materials studied. Also, the irradiation with CO₂ laser as a surface pretreatment may skip the previous acid etching, thus, smear layer is not removed, which could decrease the post-treatment dentin hypersensitivity. However, there is a need for further studies on the effects of different laser parameters and low shrinkage bulk-fill resins. Within the limitations of the present study, it could be concluded that there is no significant difference between the surface pretreatment with CO₂ laser and 37% phosphoric acid with regard to microleakage following bulk-fill resins, as demonstrated through dye penetration. Further studies are needed for the determination of other efficient, safe laser hard tissue etching parameters, filling methods and bulk-fill restorative materials.

**Ethical Considerations**

This study was approved by the Ethics Committee of the Metropolitan University of Santos (UNIMES) under the number protocol 1.741.384.

**Conflict of Interests**

The authors declare no conflict of interest.

**Acknowledgments**

The authors would like to thank Professor Maria Cristina Chavantes and Dr. Alvaro Pereira de Oliveira who generously loaned us the Ultralase the 30 ultra-short pulsed CO₂ laser (South Inc., USA). The authors also would like to thank the laboratory technician Márcio José Figueira Chaves who helped us with the images.

**References**


