

Effect of Conventional and Resin-modified Glass-Ionomer Liner on Dentin Adhesive Interface of Class I Cavity Walls After Thermocycling

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Clinical Relevance

The long-term quality of a dentin adhesive interface appears to be maintained when a resin-modified glass-ionomer liner is used.

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SUMMARY

Objective: The aim of this *in vitro* study was to analyze the effect of glass-ionomer cement as a liner on the dentin/resin adhesive interface of lateral walls of occlusal restorations after thermocycling.

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Materials and Methods: Occlusal cavities were prepared in 60 human molars, divided into six groups: no liner (1 and 4); glass-ionomer cement (GIC, Ketac Molar Easymix, 3M ESPE) (2 and 5); and resin-modified glass-ionomer cement (RMGIC, Vitrebond, 3M ESPE) (3 and 6). Resin composite (Filtek Z250, 3M ESPE) was placed after application of an adhesive system (Adper Single Bond 2, 3M ESPE) that was mixed with a fluorescent reagent (Rhodamine B) to allow confocal microscopy analysis. Specimens of groups 4, 5 and 6 were thermocycled (5°C-55°C) with a dwell time of 30 seconds for 5000 cycles. After this period, teeth were sectioned in approximately 0.8-mm slices. One slice of each tooth was randomly selected for confocal microscopy analysis. The other slices were sectioned into 0.8 mm × 0.8 mm beams, which were submitted to microtensile testing (MPa). Data were analyzed using two-way ANOVA and Tukey test ($p < 0.05$).

Results: There was no detected statistical difference on bond strength among groups ($\alpha < 0.05$). Confocal microscopy analysis showed a higher mean gap size in group 4 (12.5 μm) and a higher percentage of marginal gaps in the thermocycled groups. The RMGIC liner groups showed the lowest percentage of marginal gaps.

Conclusions: Lining with RMGIC resulted in less gap formation at the dentin/resin adhesive interface after artificial aging. RMGIC or GIC liners did not alter the microtensile bond strength of adhesive system/resin composite to dentin on the lateral walls of Class I restorations.

INTRODUCTION

Despite extensive research showing significant improvements in the clinical performance of resin-based restorative materials, polymerization contraction still remains a clinical problem. Contraction stress developed during polymerization is transferred to the tooth/resin composite interface,^{1,2} and it may cause marginal leakage, postoperative sensitivity, marginal discoloration, and recurrent caries.³⁻⁹

The use of bonding agents counteracts polymerization contraction to a certain extent, but immediate microleakage and gap formation at the tooth/restoration interface is an adverse phenomenon that occurs in different clinical situations. Adhesive

systems cannot totally prevent gap formation and microleakage as long as polymerization contraction and dimensional changes of resin composites are not substantially reduced.^{1,2}

Different methods are used to improve cavity sealing of resin composite restorations such as modulated photopolymerization,^{3,4} incremental placement techniques,^{5,6,10,11} and the use of a liner with a relatively low modulus of elasticity as an intermediary layer between resin composite and tooth (eg, glass-ionomer cement), acting as a shrinkage stress absorber.¹²⁻¹⁵

One of the most important criteria for clinical success of composite restorative materials is the effectiveness and durability of the bonded interface. While in the oral environment, composite restorative materials are submitted to different types of challenges such as thermal stress.

For all of the above mentioned reasons, the aim of this *in vitro* study was to analyze the effect of glass-ionomer cement liners on the dentin adhesive interface of occlusal restorations, with or without thermocycling. The hypothesis tested was that the use of glass-ionomer cement (GIC) and resin-modified glass-ionomer cement (RMGIC) liners does not affect dentin adhesive strength and gap formation after thermocycling.

MATERIALS AND METHODS

Specimen Preparation

Materials used in this study are listed in Table 1. Sixty freshly extracted noncarious third molars were obtained under a protocol approved by the Ethics Committee of the Bauru School of Dentistry, University of São Paulo (117/2007-FOB).

An occlusal cavity (5×3 mm) was prepared in each tooth with the pulpal floor at a depth of 4.5 mm into dentin, using a high-speed handpiece with a cylindrical carbide bur (56; KG Sorensen, São Paulo, Brazil). Following these procedures, specimens were randomly assigned to six groups (n=10). The specimens were divided into groups as follows:

- G1: No liner and no thermocycling (control)
- G2: Glass-ionomer cement liner (Ketac Molar Easymix, 3M ESPE Dental Products, St Paul, MN, USA) and no thermocycling
- G3: Resin-modified glass-ionomer cement liner (Vitrebond, 3M ESPE Dental Products) and no thermocycling
- G4: No liner, thermocycling

Table 1: *Materials Used in This Study*

Materials	Composition	Batch	Manufacturer
Filtek Z250 Universal Restorative - Shade A3	Organic matrix: Bis-GMA (bisphenylglycidyl dimethacrylate) UDMA (urethane dimethacrylate) Bis-EMA (ethoxylated bisphenol-A dimethacrylate) Camphorquinone Filler: Zirconia/silica (82% by weight, 60% by volume; average particle size 0.6 µm)	8MX	3M ESPE Dental Products, St Paul, MN, USA
Adper Single Bond 2	Bis-GMA HEMA Diurethane dimethacrylate Polyalkenoic acid copolymer Camphorquinone Water Ethanol Glycerol 1,3 dimethacrylate Silica (5 nm; 10% by weight)	6HF	3M ESPE Dental Products, St. Paul, MN, USA
Gel Dental Conditioner	37% Phosphoric acid gel	900886	Dentsply International, York, PA, USA
Rhodamine B	$C_{28}H_{31}ClN_2O_3$	33907062	Sigma-Aldrich Chemie GmbH, Gillingham, UK
Vitrebond	Powder: Glass fiber Diphenyliodonium chloride Liquid: Copolymer of acrylic and itaconic acid Water 2-Hydroxyethyl methacrylate	8ML	3M ESPE Dental Products, St. Paul, MN, USA
Ketac Molar Easymix	Powder: Glass Fiber Polyacrylic acid Liquid: Water Polycarbonic acid Polyethylene Tartaric acid	308161	3M ESPE Dental Products, St. Paul, MN, USA

- G5: Glass-ionomer cement liner (Ketac Molar Easymix, 3M ESPE Dental Products) and thermocycling
- G6: Resin-modified glass-ionomer cement liner (Vitrebond, 3M ESPE Dental Products) and thermocycling

Specimens in groups 2, 3, 5 and 6 were lined with a 0.5-mm ionomer cement liner on the pulpal floor. In order to be sure that lateral walls were free of ionomer cement, a specific syringe system, the Centrix system, (Centrix, C-R syringes, Shelton, CT, USA.), was used for application of the resin-modified glass-ionomer cement, and a calcium

hydroxide applicator was used for application of the conventional glass-ionomer cement. The specimens that received the conventional glass-ionomer cement had their pulpal floor previously treated with its own liquid for 30 seconds which was subsequently washed out. Following the application of the liner, the cavities were bonded and filled in similar, standardized sequences with the same adhesive system (Adper Single Bond 2, 3M ESPE Dental Products) and resin composite (Filtek Z250, 3M ESPE Dental Products). Enamel and dentin were acid etched (37% phosphoric acid, Gel Dental Conditioner, Dentsply International, York, PA,

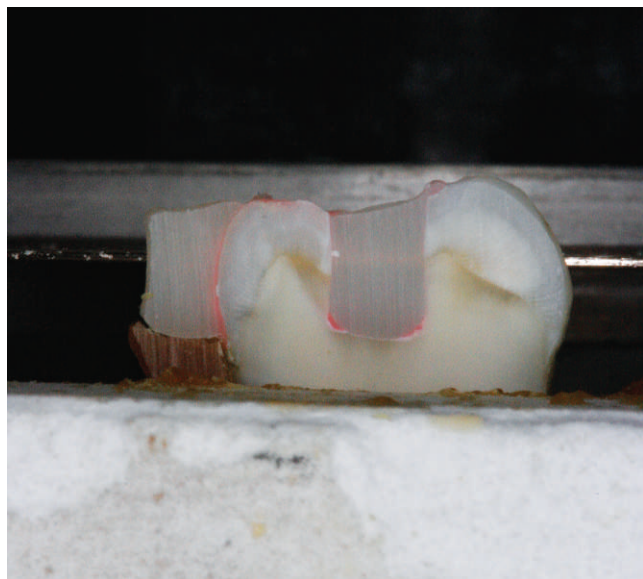


Figure 1: Specimen slice has been sectioned mesiodistally into beams. The buccal dentin/resin was kept intact for microtensile test.

USA) for 30 seconds and 15 seconds, respectively, and rinsed and dried with absorbent paper. Adper Single Bond 2 was applied in two consecutive layers and after 15 seconds, gently air dried to allow evaporation of the solvent prior to polymerization for 10 seconds with a halogen curing light (Ultralux, Dabi Atlante, Ribeirão Preto, São Paulo, Brazil) at 500 mW/cm^2 . Resin composite was applied in two horizontal increments (2-mm thickness), and each increment was light cured for 40 seconds. Specimens were stored in distilled water for 24 hours at 37°C (groups 1, 2, and 3) or submitted to a thermocycling process that included two baths (5°C - 55°C) with a dwell time of 30 seconds for 5000 cycles (groups 4, 5, and 6), before preparation for microtensile testing.

The specimens were cut buccolingually into 0.8-mm thick slices (Figure 1) parallel to the tooth long axis using an ISOMET Low Speed Saw (Buehler, Lake Bluff, IL, USA) and a diamond disk (Extex Corp., Enfield, CT, USA). One slice of each tooth was randomly selected to be analyzed via confocal microscopy (Leica TCS SPE, Mannheim, Germany, DMI 4000B with diode laser excitation of 532 nm) to verify gap formation on the adhesive interface of the buccal wall. To allow confocal microscopy analysis, Adper Single Bond 2 was mixed with a fluorescent reagent (Rhodamine B, Sigma-Aldrich Chemie GmbH, Gillingham, UK), in a concentration of 0.16 mg/mL .¹⁶

The remaining slices were sectioned mesiodistally into beams with a cross-sectional area of approxi-

mately 0.8 mm^2 , and the buccal dentin/resin interface was kept intact for the test (Figure 1).

The microtensile test was done on intact beams using a custom-made testing apparatus (Geraldelli's device) mounted in a universal testing machine (EMIC DL 500 BF, São José dos Pinhais, Brazil). Specimens were loaded at a crosshead speed of 0.5 mm/min until failure.

Both surfaces of each fracture site were observed under a digital microscope DinoLite Plus (AnMo Electronics Corporation, Hsinchu, China) with $40\times$ magnification. The fracture modes were classified as cohesive failure in resin composite, cohesive failure in dentin, adhesive failure, and mixed failure. Mean microtensile strength values were expressed in MPa.

Statistical Analysis

The data from the microtensile test was evaluated for statistical significance by two-way analysis of variance (ANOVA) ($\alpha=0.05$). Gap presence was analyzed by Fisher test ($\alpha=0.05$) and gap size by Kruskal-Wallis test ($\alpha=0.05$).

RESULTS

Results are shown in Table 2. The mean microtensile bond strength to dentin showed that the use of glass-ionomer liners did not improve the bond strength of resin composite to buccal dentin walls, with or without thermocycling. Confocal microscopy analysis showed presence of gaps in all six groups, with no statistical difference among groups, and the comparison between groups with no liner or with glass-ionomer cement liner showed an increase of gaps after thermocycling. The groups with resin-modified glass-ionomer cement liner showed no statistical difference before or after thermocycling. However, the thermocycling process seems to increase the size of gaps on the groups with glass-ionomer cement.

Figures 2 through 5 illustrate the confocal microscopy images analyzed during the study. Figure 2 shows the absence of gaps (G1 specimen), Figure 3 shows the presence and size of a gap (G1 specimen), Figure 4 shows the absence of gaps (G5 specimen), and Figure 5 shows the presence and size of a gap (G5 specimen).

The distribution of the types of failures can be seen in Table 3.

DISCUSSION

Polymerization contraction stresses are one of the shortcomings of the curing pattern of light-cured

Table 2: Mean (Standard Deviations) of Microtensile Bond Strength to Dentin, Percentage of Gaps, and Mean Size of Internal Gaps

Groups	Mean Microtensile Bond Strength, MPa	Gap Formation Presence of Gaps, %	Mean Size of Internal Gaps, μm
G1	19.28 (9.30) ^{aA}	30 ^a	1.40 (2.27) ^a
G2	16.29 (6.22) ^a	25 ^a	2.88 (6.66) ^a
G3	15.95 (7.26) ^a	25 ^a	6.63 (16.81) ^a
G4	19.74 (9.99) ^a	53 ^b	4.20 (5.23) ^a
G5	16.58 (8.61) ^a	70 ^c	12.50 (13.00) ^b
G6	16.01 (6.95) ^a	30 ^a	5.40 (8.69) ^a

Different letters indicate statistical differences among rows in the same column ($p < 0.05$).

resin composites, which may compromise the achievement of a perfect seal at the cavity wall.^{1,2} Polymerization shrinkage of a resin composite can create contraction forces that may disrupt the bond to cavity walls.^{1,2} This competition between mechanical stress in polymerizing resin composites and the bond of adhesive resins to the walls of restorations is one of the main causes of marginal failure and subsequent microleakage observed with resin composite restorations. Several researchers^{14,17-19} have sought for techniques and materials to overcome resin composite's undesirable curing effects. The evolution of the microtensile technique has permitted the evaluation of regional bond strengths in complex cavity restorations, allowing a better understanding of the bonding mechanism.²⁰⁻²²

Most dentin bond strength studies are done in free and plain surfaces with low C-factor. There are some studies that compare different C-factors with the results showing the negative potential of cavity geometry.²⁰⁻²² Microtensile tests make it possible to test bond strength on cavities walls.

Beside C-factor, measuring microtensile bond strength in cavities also involves other factors such as the type of bur used to cavity preparation, different patterns of smear layer and humidity in the different regions of a cavity, and inclination of the cavity walls at the moment of the application of the adhesive system²³ that may lead to different results than the ones obtained by adhesion on flat surfaces.

Most available adhesives show good immediate retention and adhesive interface sealing. Although after this immediate effectiveness, there is a big

concern when the adhesive dentin/resin interface is tested after aging, even after a short period of 6 months.²⁴

During aging of the adhesive interface, some phenomena lead to the degradation of the hybrid layer, especially if simplified adhesives are used. Insufficient resin impregnation in dentin, underpolymerization, separation of phases and endogenous collagenase enzyme activation are some of the factors that decrease the longevity of adhesive interfaces. In other words, clinical simplification procedures can decrease the adhesive effectiveness.²⁴

Clinical longevity related to the stability of the adhesive interface seems to involve mechanical and chemical factors such as chewing forces and the repetitive stress of contraction and expansion due to temperature changes in the oral cavity.²⁵⁻²⁸ The acid chemical agents of the dentin fluid, saliva, foods and beverages, and bacterial products are a challenge for the tooth/biomaterials interface in several ways such as unprotected collagen fibril degradation,²⁸⁻³² resinous monomer dilution (probably caused by underpolymerization)³³⁻³⁵ and resin component degradation.^{27-29,36,37}

Thermal cycles to simulate an intraoral condition have been common in several *in vitro* studies. Studies of the effect of thermocycling are contradictory, but most of them show that thermal stress increases leakage.²⁵ ISO Standard TR 11450 (1994) recommends 500 cycles in water at 5°C and 55°C, but literature revealed that more thermal cycles are necessary to simulate a long-term *in vivo* adhesive

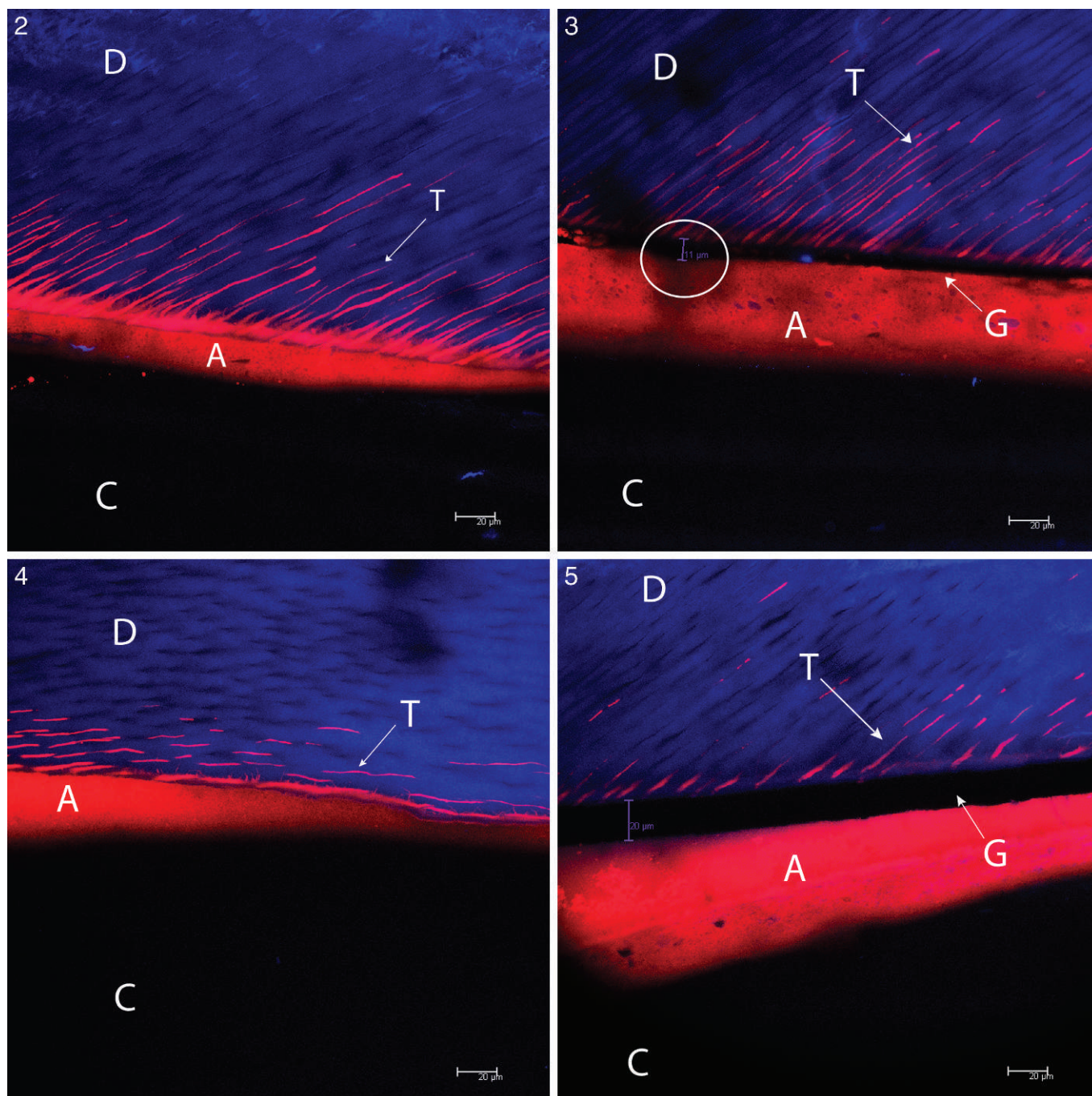


Figure 2, 3, 4 and 5: Confocal microscopy images (2) absence of gaps in G1 specimen (no liner and no thermocycling), (3) presence and size of a gap in G1 specimen, (4) absence of gap in G5 specimen (Glass-ionomer cement liner and thermocycling) and (5) presence and size of a gap in G5 specimen. (D = Dentin; A = Adhesive; C = Resin Composite; G = Gap; T = Resin tag)

stress. Wattanawongpitak and others³⁸ tested specimens with 0, 500, or 5000 thermal cycles and observed that for one of the adhesives tested, the marginal leakage significantly increased after 500 cycles, while after 5000 cycles all adhesives tested had shown a decrease in marginal integrity when compared to those with 0 cycles. There is no evidence

of the number of cycles needed to simulate an *in vivo* experience, but a provisory estimate suggested by Gale and Darvel²⁵ is that 10,000 cycles are equivalent to one year of clinical life.

An artificial aging effect induced by thermocycling is not totally established. Two mechanisms can occur: 1) hot water can speed up the hydrolysis and

Table 3: *Types of Failure (% Per Group)*

	Types of Failures - % (# of beams)			
	Adhesive	Mixed	Cohesive in Resin	Cohesive in Dentin
G1	80.95 (17)	4.76 (1)	4.76 (1)	9.53 (2)
G2	65.00 (13)	25.00 (5)	5.00 (1)	5.00 (1)
G3	71.45 (20)	14.28 (4)	7.14 (2)	7.14 (2)
G4	69.23 (18)	30.77 (8)	– (0)	– (0)
G5	78.26 (18)	17.39 (4)	4.35 (1)	– (0)
G6	76.20 (16)	9.52 (2)	9.52 (2)	4.76 (1)

decomposition of interface components, and 2) a repetitive thermal contraction and expansion stress can be generated. De Munck and others³⁹ observed that thermocycling leads to a contraction/expansion stress combined with an increase in chemical degradation.

According to some authors,⁴⁰⁻⁴³ the creation of an intermediate elastic liner between tooth and resin composite, functioning to absorb the stress associated with the shrinkage polymerization, can better be achieved with a resilient material having a lower modulus of elasticity than resin composite. Glass-ionomer cement used as a liner has been reported to improve the restoration marginal quality, increasing bond strength and decreasing gap formation at the dentin/resin composite adhesive interface.^{42,43}

The use of confocal microscopy to verify the influence of the resin-modified glass-ionomer cement and glass-ionomer cement liners and thermal aging on the dentin/resin composite interface was based on a study by Watson and Boyde⁴⁴ that described the use of confocal microscopy in dentistry. This microscopy allows the attainment of high-resolution images of fine tooth specimens that are semitransparent.⁴⁴ The specimen's surface images are produced in a similar way to those obtained with scanning electronic microscopy (SEM), but with the advantage of simplicity in technique.^{44,45}

The use of confocal microscopy images offers less image rejection due to focusing problems and better resolution than conventional imaging, especially when imaging biologic structures.⁴⁶ There is no need

to dry the specimens as for SEM and transmission electronic microscopy (TEM). These advantages lead to less risk of contraction artifacts produced by drying and allow the specimen to be studied with an additional microscopy technique.^{45,47,48}

An additional advantage of the confocal principle is to allow the visualization not only of the specimen surface but also of its subsurface.⁴⁹⁻⁵¹ Therefore, this new approach offers the possibility of obtaining a tridimensional image, leading to a more specific and informative correlation when compared to bidimensional analysis.^{44,49}

The micromorphologic evaluation of the dentin/restoration adhesive interface can be improved by incorporation of dye in adhesive agents.⁵¹ The mix of fluorochromes with different adhesive components allows characterization of the smear layer micromorphology and thickness, distribution of these components on dentin, resin tag extension, adhesive layer thickness, and defects or alterations on the restoration interface.⁵¹ There is no consensus, however, on the optimal dye or concentration to use. Another concern is the influence that the fluorescent agent can have on the polymerization of the adhesive system as well as on its properties and behavior. Depending on the dye concentration, reduction of monomer conversion and decrease of adhesive bond strength can be expected.¹⁶ Most studies do not mention the accurate concentration of mixed dye/adhesive systems, thus making the concentration that each researcher uses an arbitrary decision that can lead to marked differences between similar studies.

D'Alpino and others¹⁶ suggested an ideal concentration of Rhodamine B to be added to Adper Single Bond. The authors observed that the concentration was optimal for confocal microscopy analysis of the adhesive interface and did not interfere with photopolymerization and adhesive resistance of the adhesive system.¹⁶

The magnitude of generated stress during polymerization of resin composite on the dentin/resin adhesive interface is influenced by various factors such as material, insertion technique, cavity preparation, and the interaction among these factors.⁹ Some studies have shown that the use of resilient liners, as conventional and RMGIC, are capable of absorbing the polymerization contraction stress and, thus, improve the bond strength and decrease marginal microleakage.^{13,52-55} In contrast, Chuang and others⁵⁵ showed that the use of this fluid liner reduces the presence of bubbles in the resin

composite restoration class II interface, but it does not improve the marginal sealing.⁵⁶

The degree of gap formation depends on various factors, some related to the patient (eg. tooth and cavity) and others related to the materials (eg. adhesive systems and resin composites) and technique, as previously described. This study showed that the use of a RMGIC liner was capable of providing a better marginal quality after an artificial aging process of thermocycling. The number of gaps significantly increased after aging when no liner or GIC liner was used. Ratih and others⁵⁷ had considered that the use of a GIC could completely stop the fluid movement just after the restoration process. However, long-term degradation of GIC can affect adhesive quality and other physical properties.^{58,59} Therefore, the benefit of GIC liner on restoration sealing is still questionable.

In the present study, the specimens with conventional GIC liner, after having been submitted to the thermal stress of thermocycling, exhibited a mean gap size significantly higher than the other tested groups. However, the groups with RMGIC liner presented a lower percentage of gaps with thermal stress.

According to Wilson and Kent,⁶⁰ RMGIC is used as a liner in resin composite and amalgam restorations with the objective of protecting the restored tooth against thermal and chemical trauma and also of keeping the cavity margin sealed.

Christensen⁶¹ noticed the need of further studies to determine the long-term RMGIC behavior because of its tendency to absorb water from saliva, food, and drinks, leading to swelling as well as the hydrolysis process. According to Huang and others⁶² resin-modified glass-ionomer cement is capable of hygroscopic expansion when stored for one week in distilled water, decreasing the presence of gaps. These studies can explain the lower number of gaps on the groups with RMGIC liner, since they had remained immersed in distilled water for 24 hours (no thermocycled) or seven days (thermocycled).

This study was aimed to evaluate the influence on the dentin/resin interface of glass-ionomer and resin-modified glass-ionomer cement liners on bond strength and on internal gap formation. The analysis of these properties are in accordance with Peutzfeldt and Asmussen,⁹ concluding that there is no relation between bond strength and marginal gap formation, and suggesting that the bond strength cannot be a sole determinant factor on restoration marginal quality, *in vivo*.

CONCLUSIONS

The use of glass-ionomer and resin-modified glass-ionomer liners did not improve the immediate bond strength of a resin composite to the lateral cavity walls of occlusal restorations. RMGIC liner seems to achieve better stability of the dentin/resin composite interface after thermocycling when considering the number and size of the gaps formed.

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