Microleakage Evaluation of Class II Composite Resin Restorations with Different Thicknesses of Resin-Modified Glass Ionomer

H. Moosavi 1, H.S. Mohammadipour 2, M. Karamimoghaddam 3.

1 Associate Professor, Dental Materials Research Center, Department of Operative Dentistry, School of Dentistry, Mashhad University of Medical Sciences, Mashhad, Iran
2 Assistant Professor, Dental Materials Research Center, Department of Operative Dentistry, School of Dentistry, Mashhad University of Medical Sciences, Mashhad, Iran
3 Dentist, Private Practice, Mashhad, Iran

Abstract
Background and Aim: One of the weaknesses of Class II composite resin restorations is gingival microleakage which contributes to postoperative sensitivity and secondary caries. The aim was to evaluate the microleakage in Class II composite resin restorations with different thicknesses of resin-modified glass ionomer (RMGI).

Materials and Methods: In this in-vitro study, standardized Class II slot cavities were prepared on the proximal surfaces of 90 molars. In group 1, total-etch adhesive and composite resin were applied using the incremental technique. In group 2, total-etch adhesive and composite were applied using the bulk technique. In group 3, 1 mm of RMGI was applied over the gingival floor, which was covered with increments of composite. In group 4, 1 mm of RMGI was placed on the gingival floor and covered with composite using the bulk technique. In group 5, 2 mm of RMGI was applied over the gingival floor, followed by an incremental composite placement. In group 6, 2 mm of RMGI was placed on the gingival floor, and the cavity was filled using the bulk technique. After thermocycling and staining with methylene blue, the samples were sectioned, and the extent of dye penetration was examined under a stereomicroscope. Data were analyzed using Kruskal-Wallis test and logistic regression (α=0.05).

Results: The lowest and highest dye penetrations were observed in the first, second, and fifth groups, respectively. The RMGI thickness did not influence the microleakage scores significantly in either composite placement techniques (P=0.828).

Conclusion: None of the restorative techniques completely eliminated microleakage of Class II composite resin restorations.

Key Words: Dental Leakage, Composite Resins, Glass Ionomer, Open Sandwich

Introduction
The reliability and durability of the marginal seal are crucial for any restoration to conserve the pulpal health and to raise the longevity of the restoration [1]. One of the weaknesses of Class II composite resin restorations is the microleakage at the gingival margin of the proximal box, which contributes to postoperative sensitivity and high incidence of secondary caries, accounting for many clinically failed restorations [1]. To reduce or eliminate this problem, several composite insertion techniques, different light-curing methods, different curing models (pulse stepped curing) [2], and the use of light-guided instruments (clear matrix and reflecting wedges) [3] have been proposed in the literature. The incremental placement of composite resin using pre-cured composite inserts, beta quartz inserts [4], or
auto-polymerizing composites [5], and the recent application of bulk-fill composites with low shrinkage characteristics [6] have been suggested to reduce the polymerization shrinkage which is a causative factor of gingival gap and microleakage. The three-sided curing technique has been recommended to guide the shrinkage towards the margin rather than away from it to avoid gap formation [7]. In this method, the gingival increment is irradiated by the use of light-reflecting wedges from a cervical direction, while the buccal and lingual increments are irradiated through the respective sites [8]. Another approach that has been reported to reduce gingival microleakage is the sandwich technique. In this method, glass ionomer cements (GICs) or other materials such as flowable composite resins [9] are applied as an intermediate layer at the gingival margin, and the remainder of the cavity is filled with incremental or bulk-fill techniques [10]. Despite these efforts, microleakage is still present at the gingival margin. Unlike amalgam, traditional composites are sticky and pull away from the cavity wall when the placement instrument is withdrawn. This difficulty in achieving close cavity adaptation leads to incomplete marginal sealing [11]. To overcome these problems, bulk-fill composites have been developed by densely loading fillers into hybrid composite resins with improved mechanical properties such as decreased wear and increased packability and curing depth [10]. The reduced polymerization shrinkage achieved through increased filler loading may offer a significantly decreased marginal leakage. However, the increased viscosity and modulus of elasticity are thought to prevent complete wetting of the cavity walls during composite resin placement. Therefore, an intermediate layer of restorative material has been suggested for improving both marginal integrity and adaptation of a high viscosity composite to cavity walls [10,11]. Restorative materials advocated for this purpose include auto-polymerizing composite resins [10], flowable resin composites (FRC) [12,13], and self-cure and resin-modified GICs (RMGICs) [14,15]. RMGICs exhibit molecular bonding to dentin and enamel, bacteriostatic activity, thermal expansion similar to that of enamel and dentin, and a slow setting reaction with a low polymerization shrinkage [14]. The "open-sandwich" technique has been suggested to be used in patients who are at medium and high risks of caries; in this technique, an RMGI is applied over the gingival floor of the proximal box, extending out to the cavosurface margins. This exposure of the RMGI to the oral environment may cause surface deterioration as a result of the high solubility of the RMGI in oral fluids [16]. However, the improved mechanical and physical properties, compared with those of conventional GICs, increase the quality and longevity of open-sandwich restorations [15,17]. This technique allows the dentist to benefit from the clinical advantages of RMGIs, including tri- or dual-cure setting, fluoride release, low coefficient of thermal expansion, greater tolerance to moisture compared to composite resins, and reduced volume of resin used for restoration [14,15]. Also, the high elastic deformation or flowability of RMGIs during early setting stages can act as a stress absorber, leading to a reduced stress transfer towards the bonding interface [14]; consequently, an improved marginal seal has been reported in several studies [17-20]. The purpose of the present in-vitro study was to evaluate whether an intermediate layer of RMGI with different thicknesses would eliminate or significantly decrease microleakage at the gingival margin of Class II composite resin restorations using various placement techniques.

Materials and Methods

Ninety-three sound human extracted mandibular molars without caries, restorations, or cracks were chosen for the study, following the approval by the Commission for Medical Ethics of Mashhad University of Medical Sciences (IR.mums.sd.REC.941133). The teeth were collected during three months prior to the study. Soft tissue, calculi, and plaque were removed using rubber cups and pumice-water slurry after hand scaling instrumentation. The teeth were immersed in 1% thymol solution for one week and stored in normal saline until the experiment. Afterward, the samples were mounted in a self-cure acrylic resin (Acropars, Marlic Co., Tehran, Iran). Class II slot cavities were prepared using a high-speed handpiece and straight fissure diamond burs (ISO 806 314, Hager & Meisinger.
GmbH, Neuss, Germany) on one of the proximal surfaces of each tooth with a 3-mm buccolingual width, a 1.5-mm mesiodistal depth at the gingival margin, and an occlusogingival height of 5 mm, approximately 1 mm below the cementoenamel junction (CEJ) with the cavosurface margins as a butt joint. The diamond burs were replaced after every five preparations.

Prior to restoration, each tooth was wrapped with a matrix band using a Tofflemire retainer. All the cavity preparations and restorations were performed by a single expert operator. The teeth were then divided into six test groups (n=15). The restoration of the prepared teeth was performed as follows:

**Group 1:** The cavities were etched using 35% phosphoric acid (Ultra-Etch, Ultradent Products Inc., South Jordan, UT, USA) for 20 seconds at enamel margins and for 15 seconds in dentin substrate, rinsed with a copious amount of water for 10 seconds to remove the remnant of the etchant and then air dried. An ethanol/water-based adhesive (Adper Single Bond 2; 3M ESPE, St. Paul, MN, USA) was then applied to the etched surfaces according to the manufacturer’s recommendations. Two consecutive layers were applied, and the solvent was gently air-dried and then light-cured for 10 seconds with a light-curing device (Bluephase C8, Ivoclar Vivadent AG, Schaan, Liechtenstein, Germany) at a light intensity of 650 mW/cm². The light intensity was checked after every five restorations. Next, the samples were restored using a universal microhybrid composite (Filtek™ Z250; 3M ESPE, St. Paul, MN, USA) via the horizontal incremental technique; the first increment was placed on the gingival floor with a 1-mm thickness, and the rest of the cavity was filled with two layers of 2-mm thickness. Each increment was cured for 20 seconds from the occlusal aspect.

**Group 2:** In this group, a posterior bulk-fill composite resin (Tetric® N-Ceram Bulk-Fill; Ivoclar Vivadent AG, Schaan, Liechtenstein, Germany) was used in one layer for the restoration of the cavities through the bulk-fill technique and then light-cured for 20 seconds from the occlusal aspect. The etching and bonding procedures before restoration were the same as group 1.

**Group 3:** An RMGI (GC light-cure universal restorative; GC Corp., Tokyo, Japan) was mixed according to the manufacturer’s recommendations and was placed on the gingival floor of the cavity to a thickness of approximately 1 mm and light-cured for 20 seconds. The thickness was evaluated by a standard William’s periodontal probe, according to the original cavity depth. The remainder of the cavity was then etched, bonded, and restored with Filtek™ Z250 according to the protocol followed for group 1.

**Group 4:** All procedures for restoration in this group were the same as group 3, except for using Tetric® N-Ceram Bulk-Fill composite resin for the restoration of the cavities in one layer of 4-mm thickness. The composite resin was cured for 20 seconds from the occlusal aspect.

**Group 5:** The bonding procedures were the same as group 3; however, two layers of RMGI with a 1-mm thickness were applied. Each layer was cured for 20 seconds and then prepared for Filtek™ Z250 composite resin insertion according to the protocol followed for group 1.

**Group 6:** After applying two layers of RMGI with a 1-mm thickness and light-curing of 20 seconds for each layer, Tetric® N-Ceram Bulk-Fill was applied in one layer for restoring the rest of the cavity, followed by light-curing. In all groups, after removing the matrix band, the restorations were light-cured from the buccal and lingual aspects for 20 seconds. After matrix removal, the completed restorations were finished and polished using a fine diamond point and a series of abrasive discs (Kerr Corp., Orange, NJ, USA). The teeth were then placed in isotonic saline in a water bath at 37°C for 24 hours and thermocycled for 1000 cycles at a temperature range of 5°C to 60±5°C with a dwell time of 20 seconds for each temperature.

For microleakage evaluation, the root apices of the teeth were sealed with sticky wax, and all the surfaces were coated with two layers of nail varnish to 1 mm beyond the gingival margins of the restorations. The teeth were soaked in 0.5% methylene blue dye solution in a 37°C water bath for 48 hours. After removal of the dye solution, the samples were thoroughly washed under tap water and sectioned mesiodistally into halves along their long axes using a diamond disc (CNC Machine, Nemo Phanavaran Pars Co., Mashhad, 2018; Vol. 30, No. 3
Iran) with water coolant. The sections were blindly examined for dye penetration by another evaluator using a stereomicroscope (Dino-Lite Pro, Anmo Electronics Corp., Taiwan). The cervical marginal microleakage was recorded according to the following criteria [9]:

Score 0 = No dye penetration
Score 1 = Dye penetration limited to enamel
Score 2 = Dye penetration beyond the dentinoenamel junction (DEJ) but limited to two-thirds of the cervical wall
Score 3 = Dye penetration beyond two-thirds of the cervical wall but not to the pulpal wall
Score 4 = Dye penetration to the pulpal wall

Data related to dye penetration in the six study groups were analyzed using Kruskal-Wallis test and logistic regression. The power of statistical tests was 75%. The significance level was set at 0.05.

Results
Data related to the number and percentage of dye penetration in the studied groups are presented in Table 1.

None of the groups showed complete prevention of dye penetration. The descriptive results indicated the best marginal sealing as well as the lowest dye penetration in the first and second groups (conventional and bulk-fill composite resins with no RMGI, respectively). The highest microleakage was noted in the fifth group (2 mm of RMGI in combination with the conventional composite resin). However, Kruskal-Wallis test showed no significant difference in the microleakage scores among the experimental groups (P=0.828, X²=2.147, df=5). The P-value indicated homogeneity among the six experimental groups. The logistic regression model showed no significant association between the RMGI thickness (P=0.538) and the composite resin placement technique (P=1.00) in terms of the dye penetration score. The frequency distribution of dye penetration at the tooth-restoration interface based on the dye penetration scores is presented in Diagram 1.

Discussion
To date, there have been no dental restorative materials or techniques that can completely eliminate the gap and microleakage at the tooth-restoration interface [21]. Several modifications in the placement techniques or curing methods of composite resins might improve the marginal seal, including the application of resins with a low polymerization shrinkage such as bulk-fill composite resins [6], the incremental technique of composite resin insertion, and the use of RMGIs or flowable liners [9] under composite resin restorations.

In the current study, a dye penetration test was used to assess the marginal seal and microleakage. One of the most frequently used methods for a simple evaluation of microleakage with low expenses is the dye penetration test which provides quantitative and comparable results [22]. However, this method has some limitations including the subjectivity of readings as well as overestimation due to the low molecular weight of the dye; these shortcomings have limited the application of this test for sealing measurements [23].

The outcome of the present study indicated that the first and second groups, which were restored with conventional and bulk-fill composite resins, respectively, exhibited a lower microleakage in comparison with the groups with the RMGI liner. However, the difference among the groups was not significant. This outcome was confirmed by a previous study performed by Shafiei and Akbarian [24] who showed superior marginal sealing with total bonding compared to the open-sandwich technique with nanofilled RMGI.

In the current study, the low microleakage in the experimental groups with no RMGI liner may be related to the adhesive nature of the dentin bonding agent that was used in these groups. The Adper Single Bond 2 is a water/alcohol-based adhesive system that contains a polyalkenoic acid copolymer derived from the GI chemical bonding concept [25]. In addition to the micromechanical retention, this bonding agent can chemically adhere to dental structures. It has been reported that the polyalkenoic acid copolymer forms calcium-polyalkenoate complexes at the superficial 3-µm layer of dentinal tubules. Furthermore, when water-containing systems are applied to the air-dried dentin, they plasticize the collapsed collagen by their water content, which may gradually be expanded again to facilitate the
infiltration of resin monomers [25]. These developments combined with the thick adhesive layer produced by this particle-filled adhesive might stabilize the bonded interface due to water stability and reduced stress [25].

One of the purposes of the use of RMGI liners under composite resin restorations is to benefit from the stress buffering capacity of these materials to resist the debonding stress during polymerization shrinkage. It seems that these benefits can be provided by particle-filled dentin bonding agents. This study could not demonstrate the advantages of the application of RMGIs at the gingival margin of Class II composite resin restorations for reducing or eliminating the marginal gap and microleakage. Those samples that contained an RMGI liner, applied according to the open-sandwich technique, presented more dye penetration than the samples without an RMGI lining. The results of the current study were in contrast to the findings of Kasraei et al [11] as they showed that the use of an RMGI as a liner in Class II composite resin restorations significantly decreases the microleakage. In spite of the application of Adper Single Bond 2 adhesive in the groups with no liner, they applied the RMGI using the closed-sandwich technique in contrast to the open-sandwich method that was used in the present study. Nevertheless, the open-sandwich technique creates additional interfaces exposed to the oral environment where the fluoride ions can release from the ionomer materials near the tooth-restoration interface; this is of great importance, especially in patients with high caries incidence.

The findings of a study by Karaman and Ozgunaltay [26] also contradicted the results of the present study. They stated that the placement of an RMGI liner reduces the microleakage of the studied composite resins [26]. In a study by Chuang et al [27], the composite resin restorations lined with an RMGI presented superior marginal sealing in moderate-sized and deep cavities. Therefore, it seems that the benefit of using RMGI liners under composite resin restorations for reducing polymerization shrinkage and microleakage still remains controversial.

The findings of our study were not in agreement with the results of the studies which showed a reduction in the cervical marginal microleakage when an RMGI intermediate layer was used [28-30]. The viscosity of the RMGI, the placement method, the operator’s skill, and using a subjective criterion (scoring) for the evaluation of dye
penetration may influence the microleakage of the open-sandwich technique [31,32].

The lack of effectiveness of RMGIs in the reduction of microleakage of composite resin restorations in the present study may be attributed to the RMGI placement method which was done using an explorer instead of the injection method. Another reason that may have reduced the effectiveness of the RMGI in the open-sandwich technique in the current study was that this material (GC light-cured universal restorative) shows a volumetric contraction upon curing due to its resinous component, which may increase the marginal gap and leakage [33].

In addition, the number of thermal cycles in the aging process can affect the amount of leakage. In the present study, 1000 cycles of thermal aging were done in contrast to 600, 250, and 100 cycles in similar studies [28-30].

According to the results, a 1-mm-thick RMGI, as an intermediate layer, showed better sealing ability than 2 mm of RMGI with no statistically significant difference. These results are in agreement with the findings of the studies in which the use of a thin liner reduced the microleakage [9,34,35]. Also, a previous study demonstrated that 1-mm-thick lining materials could compensate for the contraction stresses from the overlying composite due to viscoelastic properties [27]. Verification of the thickness of the intermediate layer in a deep proximal box is difficult in the clinical settings. In a study by Chuang et al [27], restorations lined with RMGIs presented increased internal void formation in deep cavities. It seems that the greater microleakage related to the 2-mm thickness of RMGI may be attributed to pseudo microleakage that is related to the porosity of the RMGI microstructure [27]. In other words, the leakage can be attributed to the porosities and micro-gaps within the GI structure, which facilitate dye penetration and cause overestimation of leakage.

The results of the present study were completely consistent with the outcome of a study by Majety and Pujar [17]. The similarity between these two studies may be related to the use of the same dentin bonding agent. Although the correlation between clinical evaluations and in-vitro dye penetration testing may not be documented, the latter is still a popular and valuable test as a preclinical screening method to compare the sealing ability of different adhesive materials and techniques.

There were several limitations to the current study, similar to other laboratory studies. In the present study, different pH levels and mechanical loadings were not considered to simulate the intraoral conditions and mastication forces. Since excessive variability happens in the material composition from one manufacturer to another, the results cannot be generalized to all materials. Further investigations, particularly clinical trials, might be valuable in this respect.

Conclusions
Considering the limitations of the current in-vitro study and based on the obtained results, the following conclusions were drawn:

1- There was no significant difference between incremental and bulk-fill composite resin insertion techniques with Filtek™ Z250 and Tetrix® N-Ceram Bulk-Fill, respectively, in terms of the leakage scores.

2- The RMGI, as a liner in the open-sandwich technique, in combination with conventional and bulk-fill composite resins could not decrease the microleakage at the tooth-restoration interface.

3- The difference in the thickness of RMGI layers did not significantly affect the microleakage; however, lower thicknesses caused a lower microleakage.

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