

Influence of cervical margin relocation and adhesive system on microleakage of indirect composite restorations

> **S. KÖKEN¹, J. JULOSKI¹⁻², M. FERRARI¹⁻³**

¹ Department of Prosthodontics and Dental Materials, School of Dental Medicine, University of Siena, Italy

² Clinic for Paediatric Dentistry, School of Dental Medicine, University of Belgrade, Serbia

³ Department of Restorative Dentistry, School of Dentistry, University of Leeds, Leeds, UK

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ABSTRACT

Aim The aim was to evaluate the influence of the cervical margin relocation (CMR) and the adhesive system on the microleakage of indirect composite restorations with proximal margins located below the cemento-enamel junction (CEJ).

Materials and methods Standardized MOD cavities with proximal margins located 1 mm below CEJ were prepared in 20 human molars and divided into 2 groups. Mesial margins in both groups were elevated with a flowable composite. Distal margins were not elevated. Composite CAD/CAM overlays were cemented with a resin composite; in Group 1 in combination with a universal adhesive in selective enamel etch mode, whereas in Group 2 with a three-step total-etch adhesive. Differences in leakage either at mesial or distal adhesive interface were evaluated for statistical significance ($P < 0.05$).

Results In Group 1 statistically significant differences emerged in microleakage scores between CMR and non-CMR sites; higher scores were present at CMR sites. In Group 2 no statistically significant differences existed between CMR and non-CMR margins. When the non-CMR sites were compared between the two groups, significantly lower scores were observed in Group 1 compared to Group 2.

Conclusion The CMR technique and the adhesive system employed for luting indirect restorations might represent a significant factor affecting microleakage at the interface below CEJ.

KEYWORDS Cervical margin relocation; Indirect restorations; Marginal seal; Proximal box elevation; Universal adhesive.

INTRODUCTION

Over the last two decades the use of adhesive restorative materials has aesthetically improved the possibilities for dental treatments in the posterior region (1–6). Minimally invasive restorations have gradually replaced conventional amalgam restorations, providing protection of intact tooth structure without sacrificing sound tissue for mechanical retention (7). In small and medium sized Class I and Class II cavities direct composite restorations are indicated and are found to be effective (8, 9). However, when the size of the cavity enlarges, the risk of polymerization shrinkage increases, which may result in marginal adaptation problems, such as fracture of adhesively formed tooth-restoration interface and microleakage (10, 11), possibly leading to postoperative sensitivity, marginal staining or secondary caries (12, 13).

Considering the detrimental effects of polymerization shrinkage and the complexity of placing a direct composite restoration in large posterior cavities, semidirect (14, 15) and indirect (16) restorations were proposed in such cases. Extraorally fabricated restorations that are adhesively cemented are less affected by polymerization shrinkage due to the reduced thickness of the resin to be cured. However, the problem that may often occur is the margin of the proximal box of indirect restorations located below the surrounding gingival margin and close to or below the cemento-enamel junction (CEJ). Impression taking, as well as the adhesive luting procedures are therefore hampered by subgingivally positioned margins. Although the surgical crown lengthening could be performed to overcome those clinical difficulties, elevating cervical margins coronally by adhesively bonding of a small amount of resin composite material on the proximal margin was proposed as an alternative to surgical procedures (17). Cervical margin relocation was first proposed by Dietschi et al. (18), and although it is well known among the clinicians there is a scarce scientific literature about

this technique (19). The subgingival location of margins and the absence of enamel at the cervical margin create the weak area for reliable bonding. Bonding to dentin is a sensitive technique and not as stable as that to enamel (20). Due to the degradation of the resin composite material and its adhesive interfaces over time under continuous occlusal loading (21), restoration margins in dentin could be more susceptible to microleakage and bacterial biofilm penetration, possibly leading to hypersensitivity or secondary caries (22).

The aim of this *in vitro* study was to evaluate the influence of the cervical margin relocation and the adhesive system on the microleakage of indirect composite restorations with proximal margins located below the CEJ. The three null hypotheses tested were:

1. no difference would be found in the marginal sealing between the proximal margins with and without CMR when a universal adhesive was used for composite onlay cementation;
2. no difference would be found in the marginal sealing between the proximal margins with and without CMR when a 3-step total-etch adhesive was used for composite onlay cementation;
3. no differences would be found in the marginal sealing between the two different adhesive systems used for composite onlay cementation when CMR was not performed.

MATERIALS AND METHODS

Teeth preparation

Twenty intact, healthy, similar sized human third molars extracted for therapeutic reasons without any visible cracks, cavities or restorations were selected for the study after informed consent of the patients was obtained. Teeth were mechanically cleaned with hand scalers, brushed with pumice and stored in 0.1% thymol solution for no longer than three months. Standardized MOD cavity preparations were created using water-cooled diamond burs (Komet Burs Expert Set 4562/4562ST, Komet, Lemgo, Germany) mounted on a high-speed handpiece. The remaining axial walls had 2 mm of thickness, and they were reduced for a cuspal coverage. Proximal, box-shaped preparations were made 2 mm in the mesio-distal and 5 mm in bucco-lingual direction. The inner angles of the cavities were rounded, and the margins were not beveled. Proximal margins in both mesial and distal sides were located 1 mm below the CEJ. Teeth were randomly assigned to two equal groups of 10 specimens each, as follows (Fig. 1, Table 1, 2).

- Group 1: mesial proximal margins below the CEJ were elevated with two increments of 1 mm each with G-aenial Universal Flo (GC Corp., Tokyo, Japan), which was bonded with a universal bonding agent

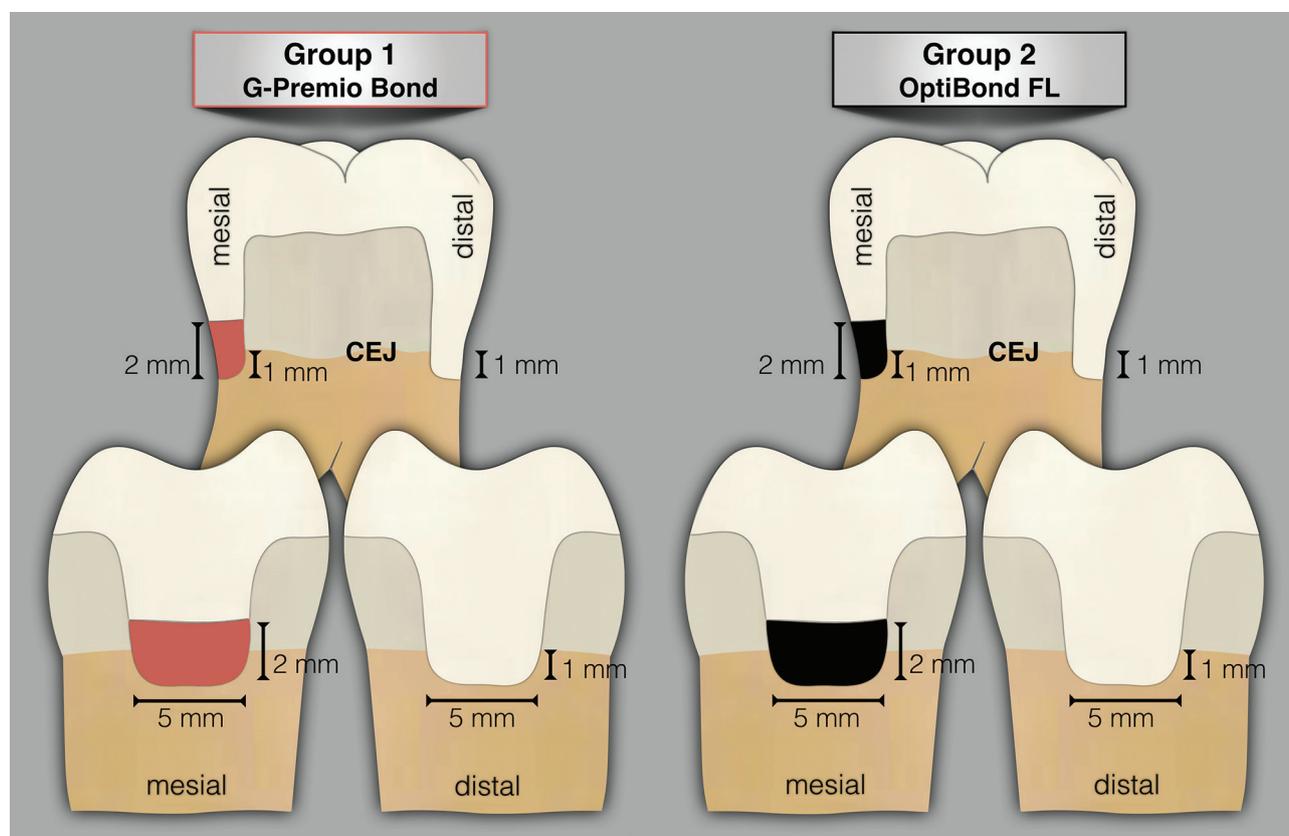


FIG. 1 Schematic description of groups.

Material (manufacturer)/Batch number	Type	Application procedure	Composition
OptiBond FL (Kerr; Orange, CA, USA) lot:5866021	Three-step etch-and-rinse adhesive	Etch-and-rinse: 37% phosphoric acid, 30 s enamel, 15 s dentin, rinsing for 30 s, gently air drying for 3 s. Primer for 20 s, gently air drying for 5 s. Bonding for 20 s, gently air thinning for 3 s, and light curing for 20 s	Primer: ethyl alcohol, alkyl dimethacrylate resins and water Bonding agent: uncured methacrylate ester, monomers, triethylene, glycol, dimethacrylate, ytterbium trifluoride, inert mineral fillers, photoinitiators and stabilizers
G-Premio BOND (GC Corporation, Tokyo, Japan) lot:1703272	Universal adhesive	Selective etching of enamel for 15 s Rinsing for 15 s Air blowing (max pressure) for 10 s Light curing for 20 s	MDP, 4-MET, MDTP, dimethacrylate monomers, acetone, water, silicon dioxide, photoinitiators
G-ænial Universal Flo (GC Corporation Tokyo, Japan) lot:1506131	High filled flowable resin composite	Each layer is light cured for 20 s	UDMA, bis-EMA, dimethacrylate monomers, silicon dioxide, fillers, pigments, photoinitiators
G-CEM LinkForce (GC Corporation, Tokyo, Japan) lot:1608231	Dual-cure adhesive luting cement	Mixture is applied on restoration's inner surface and preparation surface. Overlays are firmly pressed. Each axial wall is light cured 60 s	Paste A :UDMA, bis-GMA, dimethacrylate monomers, fillers, pigments, photoinitiators Paste B :UDMA, bis-EMA, dimethacrylate monomers, fillers, photoinitiators
G-Multi Primer (GC Corporation, Tokyo, Japan) lot:1703231	Primer for glass ceramics, hybrid ceramics, zirconia, alumina, composites, metal bonding	Applied with a micro brush on restoration's inner surface	Ethanol, Phosphoric ester monomer, γ -Methacryloxypropyl trimethoxysilane, Methacrylate monomer
GC Etchant (GC Corporation, Tokyo, Japan) lot:1610271	Etching gel %37 phosphoric acid	Selective etching of enamel for 15 s	Phosphoric acid (37%), silicon dioxide, colorant
GC Cerasmart (GC Corporation, Tokyo, Japan) lot:1609082	Force absorbing hybrid ceramic CAD/CAM block	Sandblasting and silanization of the inner surface.	Raw materials of the pre-cured composite block: UDMA, dimethacrylate monomers, bis-EMA, silicone dioxide, barium glass powder, pigments, initiator

TABLE 1 Chemical compositions and application procedures of the tested materials.

Groups	Restorative material for CMR	Adhesive system for CMR	Restorative material for overlay	Adhesive system for luting	Resin cement
Group 1: G-Premio Bond	GC G-ænial Universal Flo A2	GC G-Premio Bond	GC Cerasmart	GC G-Premio Bond	G-Cem LinkForce
Group 2: Optibond FL	GC G-ænial Universal Flo A2	GC G-Premio Bond	GC Cerasmart	Kerr Optibond FL	G-Cem LinkForce

TABLE 2 Description of the experimental groups.

G-Premio Bond (GC Corp.). Resin composite overlays were luted with a resin cement (G-Cem LinkForce, GC Corp.) in combination with the same universal bonding agent G-Premio Bond in selective enamel etch mode.

- Group 2: mesial proximal margins below the CEJ were elevated with two increments of 1 mm each with G-ænial Universal Flo (GC Corp.), which was bonded with a universal bonding agent G-Premio Bond (GC Corp.). Resin composite overlays were luted with the same resin cement (G-Cem LinkForce, GC Corp.), but in combination with a gold standard 3-step total-etch bonding system (Optibond FL, Kerr, Orange, CA, USA).

Kerr 2181 Adapt® SuperCap® matrices in steel (0.038, 5.0 mm high) were used to create the cervical marginal elevation. The circumferential matrix was carefully adjusted to eliminate the risk of overhanging of the composite material on the margins and 2 mm space is marked on the inner side of the matrix to avoid overfilling of the proximal box. Distal proximal margins were not elevated in any of the samples. To achieve CMR of mesial proximal margins and immediate dentin sealing (IDS) an universal adhesive G-Premio Bond was used in selective enamel etch mode. Enamel was etched for 15 s and rinsed for 15 s under laminar water flow. The cavity was gently dried, G-Premio Bond was applied with microbrush for 20 s, air blown with maximum pressure for 10 s and light cured for 20 s (BA Optima 10, B.A. International Ltd, Northampton, UK). In all specimens the cervical margins on the mesial sides were filled with two 1-mm increments of flowable composite G-ænial Universal Flo and the adaptation was performed with flowability of the material itself and with a ball ended hand instruments and a micro brush. Special care was taken not to layer the composite more than 2 mm in thickness. Water-cooled diamond burs (Komet Burs Expert Set 4562/4562ST, Komet, Lemgo, Germany) on high-speed handpiece were used to give final shape of each cavity after CMR was performed.

Impression

Extraoral scanner GC Aadvia Lab Scanner (GC Corp.) was used for making digital impressions of the prepared teeth. Scanned files were sent to Milling Center (GC Leuven) to create resin composite overlays (Cerasmart, GC Corp.). Teeth were kept in fresh water for two weeks at room temperature until the overlays were luted. The fit of overlays was examined under digital microscope (Nikon Shuttle Pix, Tokyo, Japan) and the digital photographs were taken at a 10x magnification.

Luting procedure

- Group 1: before luting, teeth were cleaned with ethanol and enamel was selectively etched for 15 s and rinsed with laminar water flow for further 15

s. Preparation surfaces were gently dried and GC G-Premio Bond was applied with microbrush for 20 s, air blown with maximum pressure for 10 s and light cured for 20 s (BA Optima 10, B.A. International Ltd, Northampton, UK).

- Group 2: before luting, teeth were cleaned with ethanol, first etchant gel was applied on enamel for 15 s and later etchant gel was applied on dentin further 15 s. All etchant gel was rinsed with laminar water flow for 30 s. Preparation surfaces were gently dried, Optibond FL Primer was applied with a light scrubbing motion using a micro brush for 20 s, and gently air dried for 5 s. Then Optibond FL Adhesive was applied on the preparation surfaces with a light scrubbing motion for 20 s using a micro brush but was only gently air thinned for 3 s and light cured at the final stage for 20 s (BA Optima 10, B.A. International Ltd, Northampton, UK).

Cerasmart onlays were sandblasted at approximately 3 bar pressure with 50-µm aluminum oxide particles. Subsequently, GC Multi Primer (GC Corp.) was applied to silanize the inner sandblasted surface of the overlays. Adhesive resin cement (G-Cem LinkForce GC Corp) was used to lute the overlays for both groups. G-Cem LinkForce was mixed with its special mixing tip, initial mixture was discarded on a clean paper. The latter mixture was applied on restoration's inner surface and preparation surface. The overlays were pressed firmly on teeth and the excess luting materials were cleaned with a microbrush and cotton pellets. The restoration margins were covered with a water-based glycerine gel (Airblock, DeTrey-Dentsply). Each axial wall was light cured for 60 seconds and finally occlusal surface was cured 60 s. Margins were gently finished with flexible disks (SofLex Pop-on, 3M ESPE, St.Paul, USA).

Marginal seal evaluation

All surfaces of the teeth were covered with nail varnish leaving exposed 1 mm around the area of the adhesive interfaces between the overlay and the tooth on the distal aspect and between the tooth and the CMR on the mesial aspect of the tooth. Diluted ammoniacal silver nitrate solution (1:4 ratio ammoniacal silver nitrate and distilled water) was prepared. The diluted solution was filtered using Millipore filter (0.22 nanometer filter, Carrigtwohill, County Cork, Ireland) mounted on a syringe. Each tooth was placed in a test tube with diluted ammoniacal silver nitrate solution in the presence of laboratory light. After 24 hours specimens were rinsed in water for 10 minutes three times. Nail varnish around the tooth was removed with acetone, and each tooth was placed in a test tube with the diluted photo developer solution (Kodak, Rochester, NY, 1:10 ratio photo developer solution and distilled water). After 8 hours teeth were rinsed in water three times for 10 minutes.

Each tooth was then embedded in transparent self-curing acrylic resin. Subsequently, the teeth were

sliced using a low-speed diamond saw under water-cooling (Isomet; Buehler, Lake Bluff, NY, U.S.A) into five to six 0.65-mm thick slices along their long axis and perpendicularly to the proximal margins. Samples were examined under digital microscope at 1x, 3x and 6x magnification (Nikon Shuttle Pix, Tokyo, Japan) (Fig. 2, 3). Two observers independently scored the amount of tracer along the interface as follows (23) (Fig. 4):

- score 0= no microleakage;
- score 1= 0-20% of the interface showing microleakage;
- score 2= 20-40% of the interface showing microleakage;
- score 3= 40-60% of the interface showing microleakage;
- score 4= 60-80% of the interface showing microleakage;
- score 5= 80-100% of the interface showing microleakage.

Statistical analysis

Two separate Wilcoxon signed-rank tests were performed on the data obtained from the two groups separately, in each group comparing the microleakage scores on the CMR site to those on the non-CMR site. These analyses were performed so the effect of the CMR technique on the marginal seal could be assessed for two different adhesive systems.

Additionally, in order to evaluate the influence of the adhesive system on the quality of the seal, Mann-



FIG. 2 (A) Sample of Group 1; CMR (mesial) side in which microleakage of 2 score is represented, (B) Sample of Group 1; non-CMR (distal) side in which microleakage of 1 score is represented.



FIG. 3 (A) Sample of Group 1; CMR (mesial) which microleakage of 3 score is represented, (B) Sample of Group 1; non-CMR (distal) which microleakage of 3 score is represented.

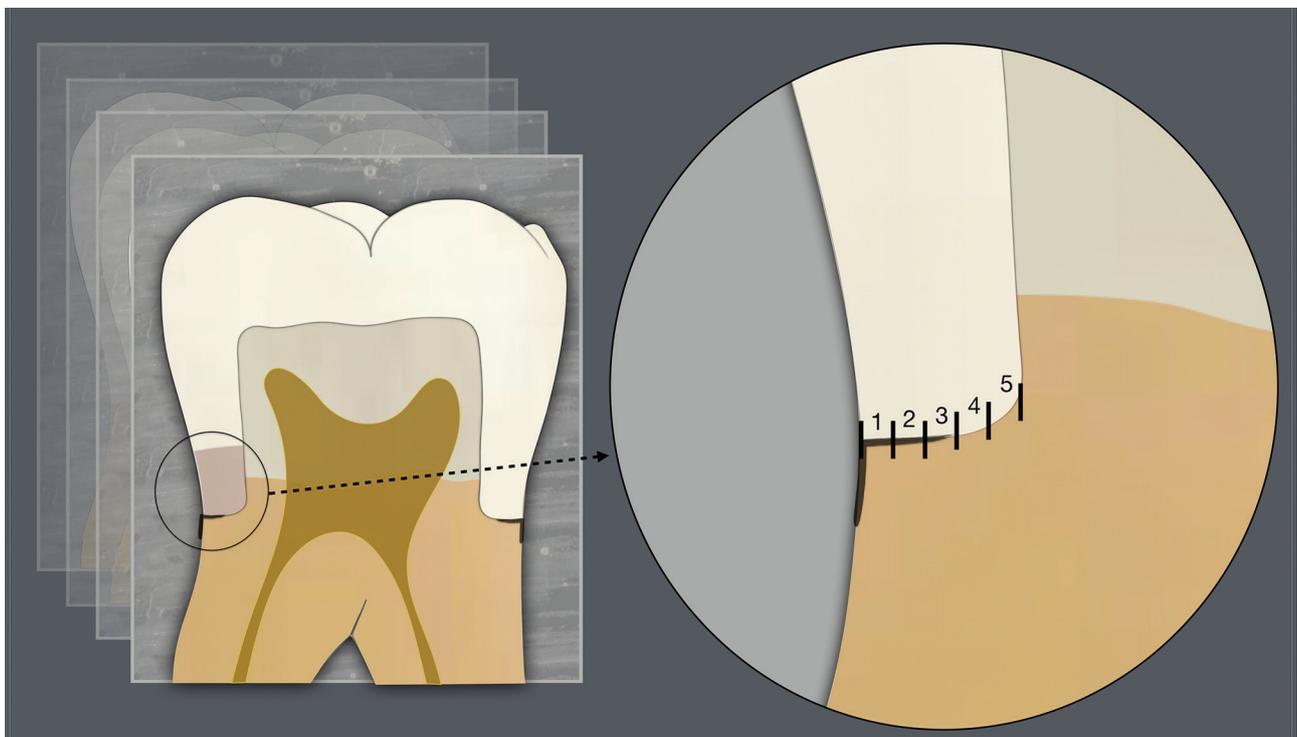


FIG. 4 The schematic drawing of the scoring system used for evaluation of the microleakage.

Whitney U test was used to assess the statistical difference in the microleakage scores between the two groups on the distal aspects of the teeth where CMR was not performed.

The significance level was set at $P < 0.05$ and the analyses were performed by means of the statistical software SPSS IBM Statistics, version 21 for Mac (SPSS Inc., Chicago, IL USA).

RESULTS

Descriptive statistics of microleakage scores are reported in Table 3, 4, 5.

Wilcoxon signed-rank test performed on data obtained from Group 1, when G-Premio Bond was used for luting the onlays, revealed that statistically significant difference existed in microleakage scores between CMR and non-CMR sites ($P = 0.000$). It showed that significantly higher microleakage was present on CMR sites (Table 3).

The same analysis conducted for Group 2 where OptiBond FL used for luting the onlays, revealed no statistically significant difference in microleakage between CMR and non-CMR margins (Table 4, Wilcoxon signed-rank test, $P = 0.491$).

Furthermore, when the non-CMR sites were compared between the two groups statistically significant differences emerged in microleakage scores (Mann-Whitney U test, P

$= 0.000$). Significantly lower scores were observed when the universal adhesive G-Premio Bond was used for luting the composite onlays directly to dentine below CEJ without CMR compared to the 3-step total-etch OptiBond FL used in the same conditions (Table 5).

DISCUSSION

The aim of this *in vitro* study was to assess the seal of the margins located in the root below CEJ when CMR technique is performed in order to relocate the margins above CEJ. Also, this paper searched for an answer to a question whether the adhesive system used for luting the indirect restoration has an impact on the leakage at the adhesive interfaces. The first null hypothesis was rejected as the results revealed that in Group 1 significantly higher microleakage was scored at the mesial aspects of the teeth where CMR was applied (median value 2) compared to the distal aspects where such treatment was not carried out (median 1). Conversely, in Group 2 no differences were detected between the two aspects of the tooth, as both median values were 2 (Table 4) and therefore the second null hypothesis was accepted. From these results it may be assumed that CMR would not be beneficial in terms of microleakage at the adhesive interface when a universal adhesive is used. On the other hand, in case of a total-etch adhesive, the results of the present study indicate

Group 1: G-Premio Bond	N	Mean	SD	Median	Interquartile range	
					25th percentile	75th percentile
CMR*	53	1,92	0,560	2,00	2,00	2,00
non CMR	53	1,16	0,758	1,00	1,00	2,00

TABLE 3 Descriptive statistics of microleakage scores recorded in Group 1. Statistically significant differences existed in microleakage between CMR and non-CMR sites (Wilcoxon signed-rank test, $P = 0.000$). Asterisk (*) indicates that significantly higher microleakage was present on CMR site.

Group 2: Optibond FL	N	Mean	SD	Median	Interquartile range	
					25th percentile	75th percentile
CMR	58	2,09	0,830	2,00	2,00	3,00
non CMR	58	2,19	0,974	2,00	2,00	3,00

TABLE 4 Descriptive statistics of microleakage scores recorded in Group 2. No statistically significant difference existed in microleakage between CMR and non-CMR sites (Wilcoxon signed-rank test, $P = 0.491$).

MICROLEAKAGE SCORE non-CMR site	N	Mean	SD	Median	Interquartile range	
					25th percentile	75th percentile
Group 1: G-Premio Bond	53	1,16	0,758	1,00	1,00	2,00
Group 2:* Optibond FL	58	2,19	0,974	2,00	2,00	3,00

TABLE 5 Descriptive statistics of microleakage scores recorded at non-CMR sites in Groups 1 and 2. Statistically significant differences existed in microleakage at non-CMR sites between Group 1 and 2 (Mann-Whitney U test, $P = 0.000$). Asterisk (*) indicates that significantly higher microleakage was observed in Group 2.

that leakage at the margins does not depend on the CMR technique, and at both proximal sites the median score value was 2. These contradictory results obtained evaluating data from Group 1 and Group 2 may suggest that the adhesive system represents a significant factor. As a matter of fact, this observation was confirmed in the third analysis, where the non-CMR sites were compared between two groups and significant differences were detected. Therefore, the third null hypothesis was also rejected.

Several papers described the clinical procedure of CMR (18, 24, 25) and several *in vitro* tests followed in order to evaluate quality of the margins after CMR was performed for reposition of the proximal margins (19). Most of the *in vitro* research investigated the marginal integrity of the relocated margins as seen under scanning electron microscopy (SEM) (26–30). A recently published paper tested the effect of CMR on the microleakage using the same method as described in the present study (31). The focus of that paper was on the type of the composite used for margin relocations and the authors reported that flowable and microhybrid resin composite show comparable performance when used for CMR prior to adhesive cementation of composite CAD/CAM overlays (31). It may be argued that the leakage test performed in both studies might be too stressing for the margins and does not truly simulate the clinical conditions. Also, it would be beneficial to further investigate whether the leakage at the adhesive interfaces recorded with *in vitro* tests corresponds to the quality of the margins as seen at SEM.

Although CMR is a well known technique and often performed among clinicians in order to avoid periodontal surgery and facilitate the restorative procedures, clinical evidence is scarce (19). Unfortunately, it is not yet pointed out what are the real clinical indications for such treatment option and in particular what are its limitations. Only 1 clinical study evaluating the CMR concept is available in the current scientific literature (32). It was shown that after 1 year of clinical service CMR was associated with statistically significant increased bleeding on probing (BoP) scores compared to shoulder preparation without CMR (32). The BoP is one of the most important clinical parameters for the evaluation of periodontal tissue health around any type of restoration and it can be directly influenced by precision of the margin, location of the margin and oral hygiene of the patient (33). The results of the present *in vitro* research could, to a certain extent, be considered in line with this clinical study, as significantly higher leakage was recorded at sites with CMR in Group 1 (Table 3). On the contrary, when another adhesive was used in Group 2 no differences between the mesial and distal margins were observed and the negative influence of the CMR could not be confirmed. Nevertheless, in this group microleakage scores recorded at the non-CMR side of the tooth were higher, which led to the absence of

difference (Table 4). Moreover, the comparison between the two adhesive systems when no prior CMR procedure was performed, clearly showed a statistically significant difference between them (Table 5). The result implies that simplified universal adhesive can be more effective in bonding to dentin compared to the most traditional total-etch adhesives, probably due to their well-known issues related to dentin bonding under tested conditions (34). Also, when used in combination with proprietary luting material for luting indirect resin restorations, less compatibility problems may occur. Another possible reason that could be of interest for clinicians is that less bonding steps may lead to less risk of mistakes and at the same time faster application.

Finally, the CMR could represent a clinical procedure that can be of a certain help for the clinicians when indirect adhesive restorations are indicated. However it must be considered that the deeper the location of the proximal margin is, the more difficult the control of the bonding steps will be (32). Also, this clinical procedure can be operator-sensitive. In order to scientifically validate the beneficial or harmful effect of CMR, more *in vitro* and *in vivo* research is needed, in particular randomized clinical trials as the most valuable source of evidence.

From the results of this *in vitro* study the following conclusions can be drawn.

1. Cervical margin relocation could influence the quality of the marginal seal of composite restorations located below CEJ. In particular, when a universal adhesive applied in a selective enamel etch mode was used for restoration luting, CMR had negative influence on the microleakage, whereas no difference existed in microleakage when a 3-step total-etch adhesive was employed.
2. The adhesive system employed for luting the indirect composite restorations represented a significant factor affecting the microleakage at the adhesive interface below CEJ when CMR was not previously performed. The universal adhesive showed better results than the three steps adhesive system under these experimental conditions.

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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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