Effect of Three Different Mouthrinses on Microleakage of Composite Resin Restorations with Two Adhesive Systems after Bleaching with 10% Carbamide Peroxide

Amir Ahmad Ajami, Mahmoud Bahari, Siavash Savadi Oskoee, Soodabeh Kimyai, Mehdi Abed Kahnamoui
Sahand Rikhtegaran, Rahim Ghaffarian

ABSTRACT

Aim: The aim was to evaluate the effects of Oral-B (OB), Listerine (LN) and Rembrandt Plus (RM) mouthrinses on microleakage of composite resin restorations bonded with two adhesive systems after bleaching with 10% carbamide peroxide.

Materials and methods: A total of 60 CI V cavities were prepared on human premolars. The occlusal and gingival margins were placed 1 mm occlusal to and apical to CEJ respectively. The teeth were randomly divided into two groups based on the adhesive system used: Excite (EX) and Clearfil SE Bond (CSE) groups. After composite resin restoration of cavities, thermocycling and bleaching with 10% carbamide peroxide for 2 hours daily for 14 days, the teeth in each adhesive group were further subdivided into three subgroups and were immersed for 12 hours in the three OB, RM and LN mouthrinses. The teeth were then placed in 2% basic fuchsin for 24 hours. After dissecting the teeth, microleakage was evaluated under a stereomicroscope at 16×. Data was analyzed with multifactor ANOVA and Bonferroni test at p < 0.05.

Results: Microleakage with EX was significantly higher than that with CSE (p = 0.009). Microleakage at gingival margins was significantly higher than that at occlusal margins (p = 0.15). Microleakage with OB was higher than that with LN (p = 0.02). However, there were no significant differences in microleakage between LN and RM (p = 1) and between RM and OB (p = 0.15). In addition, with the EX adhesive system, microleakage with OB was higher than that with LN and RM (p = 0.02).

Conclusion: In the present study, microleakage of composite resin restorations was influenced by the type of the adhesive system, mouthrinse type and the location of the cavity margin.

Clinical significance: Use of some mouthrinses, such as OB after bleaching can increase postrestoration microleakage of resin composite restorations bonded with etch-and-rinse adhesive systems.

Keywords: Laboratory research, Adhesive system, Etch-and-rinse, Self-etch, Bleaching, Microleakage, Mouthrinse.

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INTRODUCTION

Adhesive dentistry allows the use of smaller cavity preparations and preservation of tooth structure. Despite great advances in this field, there are still deficiencies in the adhesive techniques of composite resins. Probably the most important factor involved in the long-term success of composite resin restorations is microleakage, which is penetration of bacteria, liquids, molecules and ions through the tooth restoration interface. Microleakage can create clinical problems, including hypersensitivity, recurrent caries, staining of restoration margins, pulp irritation and failure of the restorative material. Therefore, prevention of microleakage is a very important consideration in the development of adhesive systems, for application in tooth restorative procedures. Historically, the first adhesive systems introduced were three-step total-etch systems, which consisted of etching, priming and bonding steps, referred to as fourth generation systems. With the passage of time advances in the adhesive dentistry resulted in the development of other adhesive systems to provide higher bond strength, sealing of the tissue permeability and more importantly ease of application. At present some of the commonly used adhesive systems belong to the fifth generation, in which the primer and the adhesive have been incorporated into one bottle, but there...
is still need for etching. During the past decade, self-etch adhesive systems have been marketed, which are referred to as the sixth generation. These systems do not need a separate etching step, and rinsing and drying steps have been eliminated; however, a separate adhesive resin or bonding agent step is necessary.3

The use of bleaching agents containing peroxide to bleach vital teeth has become very popular because of long-term clinical success and high patient satisfaction since they were introduced by Haywood and Heyman.4 Since, bleaching agents are in contact with tooth structures for a long time and there is a possibility of their inadvertent contact with dental materials, evaluation of the effect of these products on tooth structures and dental materials has attracted a lot of attention.5-7 On the other hand, preservation of restorations in the oral cavity after bleaching procedures is a great challenge in the operative and preventive dentistry. Mechanical and chemical agents, such as toothbrushes, dental floss and mouthrinses are commonly used, especially by individuals who have previously received extensive restorative procedures.8

Several in vitro and in vivo studies have evaluated the therapeutic effects of mouthrinses on plaque control.9 However, only a limited number of studies have evaluated their effect on dental materials. Recently, some clinicians have voiced concerns regarding the effect of bleaching agents on tooth-colored restorative materials.8,10,11

Since, bleaching agents function through the action of nascent oxygen species, they can exert effects on three-dimensional polymer network in composites and decrease polymerization degree12 and increase microleakage.13,14 In addition, mouthrinses, too, can induce changes in tooth structures, resulting in the same problems. For example, chlorhexidine can have an influence on the bonding procedure due to its substantivity (adsorption to tooth surfaces).15

Given the ever-increasing use of easy-to-use adhesive systems, application of different bleaching agents and procedures, and the use of various mouthrinses to promote oral health, the aim of the present study was to evaluate the effects of three different mouthrinses of Listerine (LN), Oral-B (OB) and Rembrandt plus (RM) on microleakage of Cl V composite resin restorations with two self-etch and etch-and-rinse adhesive systems after bleaching with 10% carbamide peroxide.

MATERIALS AND METHODS

In this in vitro study, 30 second human premolars extracted for orthodontic reasons were used. The teeth had mature apices and had been extracted from adolescents 1820 years of age. The teeth were free from any cracks, fractures, caries, abrasions, restorations, congenital anomalies and structural defects under visual examination and under a stereomicroscope (Nikon, SMZ 800, Tokyo, Japan). The teeth were stored in 0.5% chloramine solution until used for the purpose of the study. Before the study procedures, the teeth were cleaned of any soft tissues, debris and calculus by hand instruments and polished with a rubber cap and slurry of pumice. Class V cavities were prepared on the buccal and lingual surfaces of the teeth using an 0.8 diamond fissure bur (Diath Dental AG, Swiss Dental Instruments, CH-9435, Heerbrugg, Switzerland) in a high-speed handpiece under air and water coolant. The cavities measured 1.5 mm in depth, 2 mm in occlusogingival dimension (1 mm coronal to CEJ and 1 mm apical to CEJ) and 3 mm mesiodistally. All the cavity margins were butt-jointed without any bevels. A new bur was used after eight preparation procedures. Then the teeth were randomly divided into two groups (n = 15) based on the adhesive system used.

In group 1, Excite adhesive resin (EX) (Ivoclar-Vivadent INC, Germany) and in group 2, Clearfil SE Bond adhesive resin (CSE) (Kurary Medical Inc, Japan) were used on cavity walls according to manufacturer’s instructions. Litex 680 A light curing unit (Dentamerica, 18320 Bedford Circle, City of Industry CA 91744 USA) was used to light cure the adhesive resin according to manufacturer’s instructions. The light curing probe was 8 mm in diameter and delivered a light intensity of 600 mW/cm² perpendicular to and barely touching the surface for 10 seconds. The light intensity of the unit was tested by a radiometer (Coltolux Light Meter, Coltene/Whaledent, Altstatten, Switzerland) before the light curing procedure. A3 shade of Filtek Z250 composite resin (3M ESPE, USA) was used to restore the cavities using the incremental technique with two 1 mm thick layers. A periodontal probe was used to check the thickness of each 1 mm layer of composite resin. Each layer was light cured for 20 seconds. Restorative procedures were carried out by one operator at a room temperature of 21°C. After restorative procedures, all the samples were polished with diamond polishing burs (Diamant, Gmbh, DandZ, Goerzallee 307, 14167 Berlin, Germany) and polishing disks (Sof-lex™, 3M ESPE, Dental Products, St Paul, MN 55144, USA). Subsequently the specimens were incubated in distilled water at 37°C for 24 hours. To simulate oral conditions the specimens underwent a thermocycling procedure consisting 500 cycles at 5 ± 2°C/55 ± 2°C with a dwell time of 30 seconds and 10 seconds for specimen transfer.

Then the teeth were dried and bleached with 10% carbamide peroxide (Pola Night, SDI Limited, Bayswater, 10Victoria 3153, Australia) for 14 days for 2 hours daily.
The teeth in each adhesive group were subdivided into three subgroups of five teeth and immersed in OB, LN and RM mouthrinses for 24 hours. Then all tooth surfaces were covered with two layers of nail varnish up to 1 mm short of the restoration margins. The apices of the teeth were sealed with utility wax. The teeth were then placed in 2% basic fuchsin for 24 hours. Table 1 summarizes the composition of the materials used in the present study. After dissecting the teeth, the teeth were evaluated under a stereomicroscope (Nikon, Japan) at 16× and dye penetration at occlusal and gingival margins was classified as follows:

0: No dye penetration
I: Dye penetration up to less than half the cavity depth
II: Dye penetration more than half the cavity depth without axial wall involvement
III: Dye penetration up to the axial wall or traversing the axial wall.

DATA ANALYSIS

Data was analyzed by multifactor ANOVA. In cases in which the differences were statistically significant Bonferroni test was used for two-by-two comparisons. Statistical significance was defined at p < 0.05.

RESULTS

Microleakage scores are presented in Table 2. Analysis of data by multifactor ANOVA showed that the effects of adhesive system (p = 0.009), the type of the mouthrinse (p = 0.024) and the type of the margin (p = 0.015) on microleakage were significant. In addition, the cumulative effect of the adhesive system with the type of the mouthrinse was statistically significant (p = 0.004). However, the cumulative effects of the adhesive system-margin type (p = 0.13), mouthrinse type-margin type (p = 0.84) and adhesive system-margin type-mouth rinse type (p = 0.33) were not significant (Table 3).

Microleakage with EX was significantly higher than that with CSE (p = 0.009). Microleakage at gingival margins was significantly higher than that at the occlusal margins (p = 0.015). Two-by-two comparisons of mouthrinses with a post-hoc Bonferroni test showed significant differences in microleakage scores between LN and OB (p = 0.02), with no significant differences between LN and RM (p = 1) and between RM and OB (p = 0.15). Furthermore, microleakage of composite resins with the excite adhesive system subsequent to bleaching with 10% carbamide peroxide was higher after immersion in OB mouthwash compared to the other two mouthrinses under study (p = 0.02). However, with the CSE adhesive system

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<th>Table 1: The materials used in the present study</th>
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<td><strong>Material</strong></td>
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<td>Mouthrinses</td>
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<td>Bleaching agent</td>
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<td>Composite resin</td>
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<th>Table 2: Microleakage scores separately for each subgroup</th>
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<td><strong>Study groups</strong></td>
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no significant differences were observed in microleakage scores of the mouthrinses under study (p > 0.05) (Fig. 1).

**DISCUSSION**

*In vitro* microleakage tests provide invaluable data on the sealing ability of adhesive resins. In the present study, dye penetration technique was used for *in vitro* evaluation of microleakage at enamel and dentin margins with butt-jointed class V cavities. It is the most commonly used technique for microleakage evaluation.

Generally, gap formation and subsequent microleakage of composite resin restorations are attributed to mechanical stresses as a result of polymerization shrinkage. Factors involved in polymerization stresses are as follows in order of significance: C-factor, cavity size, the technique used to place composite resin in the cavity, the light curing technique employed, and the mechanical properties of composite resin. In the present study, efforts were made to maintain all these variables in the same level for both groups. In order to keep C-factor at the same level, all the cavities were standardized with similar sizes and shapes. Furthermore, one type of composite resin and one light-curing unit were used to restore all the cavities in both groups. In addition, to simulate oral conditions, all the specimens underwent a uniform thermocycling procedure.

The mouthrinses employed in the present study were Listerine (containing alcohol), Oral-B (alcohol-free) and Rembrandt Plus (containing H₂O₂), which represented the wide range of commercial products available on the market.

In the present study, microleakage was higher after immersion in OB compared to LN. However, unlike OB, LN contains 40% alcohol. Although alcohol-containing mouthrinses can soften Bis-GMA and UDMA-based polymers, Gurgan et al showed that alcohol is not the only agent to have a softening effect on composite resins. Yap et al showed that the effect of mouthrinses on composite resins and compomers depends on the material. Higher microleakage values with OB might be attributed to the presence of cetylpyridinium chloride (CPC) in its chemical composition. CPC is a cationic surfactant, which can reduce surface tension of the liquid and decrease intersurface tension between a solid (gap walls here) and a liquid (basic fuchsine), resulting in an increase in wetting and penetration coefficient into the capillaries (gaps here).

In the present study, etch-and-rinse (Excite) and two-step self-etch (Clearfill SE Bond) adhesive systems were used because they differ in the formation of the hybrid layer through treatment of the smear layer produced during cavity preparation. The results showed that dye penetration with CSE, which contains 10-MDP (10-methacryloxydecyl dihydrogen phosphate) is less than that with etch-and-rinse systems, which is consistent with the results of previous studies.

These findings might be attributed to differences in the chemical composition of these two adhesive systems. The clinical and laboratory efficacy of adhesives containing 10-MDP has been reported by several studies. It has been demonstrated that of the functional monomers used in the composition of self-etching systems, 10-MDP bonds to hydroxyapatite more easily and with a higher bond strength value. Hayakawa et al showed that the best bond with dentin is achieved using self-etch primers containing 10-MDP, which results in a minimum degree of dissolution.

![Error-bar graph](image)
of plaque smear and opening of dentinal tubules, reducing dentin permeability, and facilitating penetration, entanglement and polymerization of monomers with demineralized dentin.\textsuperscript{28}

According to Buonocore, the etched enamel should be completely dry and uncontaminated, and should be penetrated by a hydrophobic resin.\textsuperscript{29} Hadavi et al demonstrated that contact of etched enamel with primers (hydrophilic resins) might decrease the bond strength to enamel by 31 to 44%.\textsuperscript{30} Woronko et al suggested that water-based primers, such as those used in the composition of EX should be confined to dentin.\textsuperscript{31} However, 10-MDP contains a hydrophobic alkyl group to maintain a balance between hydrophobicity and hydrophilicity, and a double-bonded ending for effective polymerization.\textsuperscript{26,27}

Greater microleakage with EX might be attributed to the inability of the primer to access all the demineralized areas, which results in unsupported collagen fibers and ease of gap formation; it might also be attributed to residual water molecules which prevent polymerization of the adhesive system.\textsuperscript{32} In addition, EX contains HEMA, which results in a progressive decrease in water pressure, making it more difficult to replace the residual water molecules in the demineralized dentin. Furthermore, the hydrophobic monomer of Bis-GMA resists the diffusion into areas containing residual water molecules.\textsuperscript{33}

Contrary to the results of the present study, the results of one study have shown less microleakage with the etch-and-rinse systems compared to self-etch systems.\textsuperscript{34} In addition, Giachetti et al reported similar microleakage scores with the self-etch and etch-and-rinse systems at enamel and dentin margins, even after cyclic loading.\textsuperscript{35} Several studies have shown that the efficacy of adhesive systems is more dependent on operator or application protocol rather than on the chemical composition and generation classification.\textsuperscript{36,37} According to Giachetti et al self-etch adhesive systems are less sensitive to operator skills.\textsuperscript{35} In addition, these systems eliminate deteriorating variables, such as over-etching, over-wetting and over-drying and are less technique-sensitive because they have eliminated the etching and rinsing steps.\textsuperscript{38}

Another important finding of the present study was the fact that microleakage scores in both systems were higher at dential margins compared to enamel margins, consistent with the results of previous studies which have evaluated various adhesive systems,\textsuperscript{39-41} although they are inconsistent with the results of some other studies.\textsuperscript{42,43} Higher microleakage scores at dential margins compared to enamel margins might be attributed to differences in the chemical composition of these two margins. Although, enamel is a highly mineralized structure with more than 90% of its volume consisting of hydroxyapatite, dentin contains a large amount of water and organic content and has a wet surface, which makes bonding difficult.\textsuperscript{44}

Various factors are involved in differences in bonding to enamel and dentin. Bonding to enamel is a relatively easy process without any special technical requirements. In contrast, bonding to dentin is somewhat challenging.\textsuperscript{45} According to Cagidiaco et al the higher leakage observed at the cervical margin may be related to the absence of dentin tubules in the limiting 100 μm of the cervical margin and the mainly organic nature of the dentin substrate.\textsuperscript{46} Enamel, when present at the cervical margin, is usually thin, prismless and less amendable to bonding. When polymerized, the composite resin contracts toward the superior bond at the occlusal margin and away from the weaker bond at the gingival margin.\textsuperscript{42}

Based on the limitations of the present laboratory research, it is also recommended that microleakage evaluations be carried out with more accurate techniques with more cross-sections using electron microscopes for the evaluation of composite resin tooth structure interface in future studies. It is also important to consider the \textit{in vitro} nature of this study and clinically, the effects of mouthrinses on restoratives may be changed by many factors in the mouth that could not be simulated in an \textit{in vitro} study. So, it is suggested that long-term clinical studies be carried out to evaluate the effect of simultaneous use of bleaching agents and mouthrinses on microleakage of resin composite restorations bonded with different type of adhesive systems.

CONCLUSION

Based on the results of the present study, it was concluded that:

1. Microleakage scores with EX were significantly higher than those with CSE.
2. Microleakage scores at gingival margins were significantly higher than those at occlusal margins.
3. Microleakage scores with OB were higher than those with LN (p = 0.02); however, there were no significant differences between LN and RM and between RM and OB.
4. Microleakage scores of composite resin restorations bonded with EX adhesive system and bleached with 10% carbamide peroxide were higher after immersion in OB compared to those after immersion in LN and RM. However, with the CSE adhesive system no significant differences were observed in microleakage scores of the mouthrinses under study.
Effect of Three Different Mouthrinses on Microleakage of Composite Resin Restorations with Two Adhesive Systems

CLINICAL SIGNIFICANCE

Use of some mouthrinses, such as OB after bleaching, can increase postrestoration microleakage of resin composite restorations bonded with etch-and-rinse adhesive systems.

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ABOUT THE AUTHORS

Amir Ahmad Ajami
Assistant Professor, Department of Operative Dentistry, School of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran

Mahmoud Bahari (Corresponding Author)
Assistant Professor, Department of Operative Dentistry, Dental and Periodontal Research Center, School of Dentistry, Tabriz University of Medical Sciences, Gholgasht Street, Zip: 5166614713, Tabriz, Iran e-mail: bahari.dds@gmail.com

Siavash Savadi Oskoee
Associate Professor, Department of Operative Dentistry, Dental and Periodontal Research Center, School of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran

Soodabeh Kimyai
Associate Professor, Department of Operative Dentistry, School of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran

Mehdi Abed Kahnamoui
Assistant Professor, Department of Operative Dentistry, School of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran

Sahand Rikhtegaran
Assistant Professor, Department of Operative Dentistry, School of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran

Rahim Ghaffarian
General Dentist, Department of Operative Dentistry, School of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran