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In Vitro Evaluation of 2% Chlorhexidine and Glutaraldehyde Application on the Microleakage of Composite Resin Restoration in Deciduous Teeth

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Abstract

Background: Various materials are suggested for dentine rewetting, disinfection, and desensitization of a restoration cavity. Glutaraldehyde and Chlorhexidine are among the most common agents used frequently for their proven beneficial effects, such as desensitization, antibacterial, and enhanced bond strength and longevity.

Aim: The aim of this study was to evaluate the effect of separate and simultaneous application of these two agents on the microleakage of resin composite restorations of deciduous teeth.

Design: Two cavities were prepared on 52 deciduous teeth (n = 13). The samples were then randomly divided into 4 study groups according to their pre-bonding treatment. (1) Control group; (2) Glutaraldehyde treatment; (3) 2% chlorhexidine treatment and (4) simultaneous use of both Glutaraldehyde and 2% Chlorhexidine solutions. Microleakage was measured under an optic microscope with 40x magnification and was graded from 0 to 4. Statistical analysis of the results was performed using one-way ANOVA and Kruskal-Wallis tests.

Results: The difference between microleakage of the study groups was not significant ($p>0.05$).

Conclusion: Use of Glutaraldehyde and Chlorhexidine illustrates no negative effects on the microleakage of resin restorations; thus it can be presumed that using them is beneficial for desensitizing and antiseptic purposes.

Keywords

Microleakage; Glutaraldehyde; Chlorhexidine; Resin composite; Primary tooth; Deciduous tooth; In-vitro study.

Introduction

Today, dental caries is still the most common chronic childhood disease, despite all preventive efforts. Its prevalence has been reported even higher than asthma and hay fever. Caries In most cases (two-thirds) are present only at the occlusal surface of the tooth and the cumulative effects of caries with age have caused more than 95% of adults to experience caries [1].

Various treatments are performed to restore carries, including amalgam, composite, and indirect restorations. Nowadays, the use of composite restorations is more popular and has advantages such as more conservative preparation, more beauty, chemical bonding to the teeth, less heat transfer, repairability, no mercury in its composition, etc. [2]. The similarity of these restorations with the teeth is a significant point because this beauty has very good psychological effects on the patient.

In addition to the many benefits mentioned for composite restorations, these restorations also have disadvantages. Although isolation is important in all restorations, it is even more important in composite restorations, and as you know, proper isolation in children is a challenge due to the less cooperation of children. The prognosis, marginal leakage rate, and bond strength of composites have high technical sensitivity and depend on many details, including enamel and dentin etching time using light-curing time, the thickness of composite layers, etc. Composite restorations are also prone to wear, so should be limited in areas with high chewing forces [2].

One of the major disadvantages of composite restorations is microleakage after restoration. Microleakage means an undetectable clinical pathway for bacteria, fluids, chemicals, molecules and ions to pass through the surface between the restoration and the tooth [2,3]. This marginal leakage makes teeth with posterior composite resin more prone to recurrent decay than teeth with amalgam restorations [1].

Today, various methods are used to reduce the effect of microleakage. For example, the application of contemporary posterior composite resins (high volume of filler and reducing shrinkage of polymerization), and the use of enamel bevel can help. In addition, composites, bonding agents and acid etches under new developments are also useful [1]. Despite many improvements that have been made to composite systems, microleakage is still a problem.

Studies on antimicrobials such as 2% Chlorhexidine solution have been performed after acid etching (4). In many of these studies, in addition to the effect on microleakage, the bond strength has also been investigated, which usually had an inverse connection between the shear bond strength and microleakage, but this connection has not been confirmed [2,5].

In some studies, antimicrobials such as Chlorhexidine and Sodium Hypochlorite have been increased restoration microleakage [6]. Antimicrobials and disinfectants can reduce the number of bacteria in the cavity, so they are useful to prevent pulp inflammatory reactions; in the similar residual dentin thickness (RDT), the presence of bacteria in the cavity leads to more severe pulp inflammation [1]. But on the other hand, it should be noted that the use of antimicrobial solutions such as Chlorhexidine, may increase microleakage and thus increase the presence of bacteria after restoration.

In addition to the above, microleakage can cause post-restorative sensitivity [1,7]. some materials and methods have been used to reduce the sensitivity after restoration. Such as the application of Glutaraldehyde (Gluma Desensitizer®, Heraus Kulzer) [2]. Studies have also been conducted to investigate the effect of using this agent on microleakage [7]. It is important that the use of this substance, which is used to reduce one of the effects of microleakage, does not itself increase microleakage.

Silveira de Araújo et al. found dentin leakage was higher than enamel, which underscores the importance of quality adhesive systems on dentin [8]. On the other hand, thickness of deciduous teeth enamel is half thickness of permanent teeth enamel, so the importance will be higher in deciduous teeth [9].

Due to the importance of dental leakage, the controversy obtained from different researches and inadequate studies about deciduous teeth microleakage, this study evaluated the in vitro effect of Chlorhexidine and Glutaraldehyde (used separately and at the same time) on dental leakage of Composite Resin restorations.

Materials and Methods

Samples and setting

52 deciduous teeth were collected whose roots were physiologically adsorbed and exfoliated or extracted for orthodontic purposes and also had an intact crown without caries and visible cracks. After collection, the samples were washed and placed in 0.2% thymol solution (Merck, Germany) for 24 hours for disinfection, then kept in normal saline at room temperature until the study [10]. The tooth surface was cleaned with a scaling brush (additional stress such as heat from brushing on the tooth surface was avoided).

For evaluation of dentin microleakage on deciduous teeth, at first, with a diamond bur, the enamel of the tooth surface was removed on one side of the tooth, and then two class V boxes (3×1×1 mm dimensions) were prepared on the buccal and lingual surfaces of the crown of each deciduous tooth. No marginal bevel was created for the holes [11]. (Figure 1).

The cavity was prepared with 330 diamond burs with high-speed handpiece and water spray [1] and each bur was used to cut only 6 teeth [10].

The samples were randomly divided into 4 equal groups (n = 13). All cavities were etched (phosphoric acid 35%, Ultradent, USA) for 15 to 20 seconds [1] and rinsed for 20 seconds and dried to create a frosty look [10].

Four different protocols were used before filling boxes as followed:

1. Control group no need for extra preparation.
2. Treatment of the box with Glutaraldehyde (Gluma Desensitizer, Kulzer, Germany) for 45 seconds, then the cavity surface is dried, washed and re-dried [12].
3. Treatment of the box with 2% Chlorhexidine (Ultradent, USA) for 20 seconds and then drying it for 15 seconds [13].
4. First glutaraldehyde and then 2% Chlorhexidine were used, as mentioned earlier

The restoration process continued for all groups. First, the fifth generation OptiBond bonding (Kerr, Switzerland) was placed on the cavity and dried with gentle air pressure for 5 seconds [2]. It was cured for 10 seconds with a light cure device (Woodpecker model LED_D, China) with at least 600mW / cm2 light intensity and 8mm probe diameter [10]. The Palfique Asteria composite (Tokuyama, Japan) was used in the cavity and polymerized for 20 seconds.

Outcomes Measures

Samples were thermocycled (Vafaei industrial, Iran) for 1500 cycles between 5 ° C and 55 ° C baths, with 30 seconds duration for each bath and 10 seconds interval for each movement between the baths. Then, the tooth surface was covered with two layers of nail polish up to 0.5 mm left to the margin of the cavity, and the apical entrance of the pulp was sealed with bee's wax [10]. The samples were kept in 0.5% fuchsin solution (Ziehl, Iran) at 37 ° C for 24 hours [14]. The specimens were cut in half occluso-gingivaly and the dye penetration was recorded using a Loop microscope (Nikon C-PS, Japan) with 40x magnification [10] (Table 1 and Figures 1 and 2).

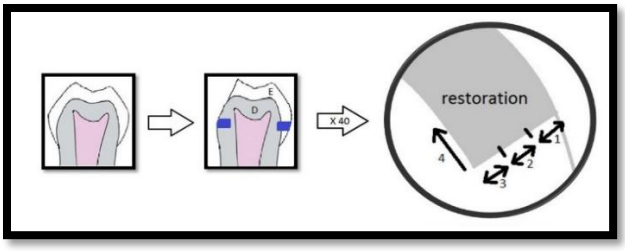

Figure 1: The boxes on enamel and dentin. Microleakage scores.

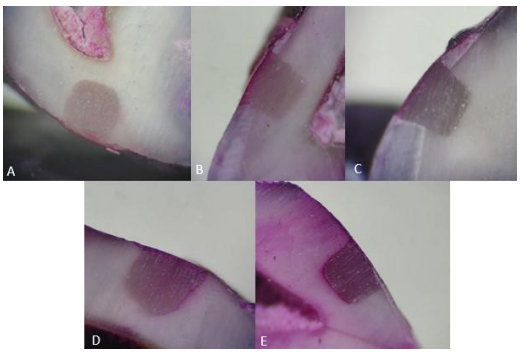

Figure 2: Microleakage scores. A: score 0. B: score 1. C: score 2. D: score 3. E: score 4.

score	Dye penetration
0	None
1	Up to one-third of the cavity depth
2	One-third to two-third of the cavity depth
3	Complete dye penetration, but not the axial wall
4	Into the axial wall

Table 1: Microleakage scores.

	Enamel				Dentin			
	Frequency	mean	Standard deviation	P-value	Frequency	mean	Standard deviation	P-value
G1 (control)	13	2.31	1.49		13	2.92	1.38	
G2 (Glutaraldehyde)	13	1.46	1.39		13	2.38	1.75	
G3 (2% Chlorhexidine)	13	1.92	1.7		13	1.77	1.69	
G4 (Glu+Chl)	13	2.23	1.64		13	2.08	1.75	
Total	52	1.98	1.55	0.59	52	2.29	1.66	0.31

Table 2: Comparison of Microleakage by type of restoration bed (enamel or dentin).

group	Frequency	Mean (Standard deviation)		P-value
		Enamel	Dentin	
G1 (control)	13	2.31 (1.49)	2.92 (1.38)	0.2
G2 (Glutaraldehyde)	13	1.46 (1.39)	2.38 (1.75)	0.22
G3 (2% Chlorhexidine)	13	1.92 (1.70)	1.77 (1.69)	0.88
G4 (Glu+Chl)	13	2.23 (1.64)	2.08 (1.75)	0.84

Table 3: Intragroup comparison of microleakage between enamel and dentin.

Each restoration cavity has two side walls (occlusal and gingival) and a floor. To determine the scores from 0 to 4, both occlusal and gingival walls were observed and the highest amount of dye penetration was considered as microleakage score.

Statistical analysis

Statistical analysis of the results was performed using one-way ANOVA and Kruskal-Wallis tests in the IBM SPSS statics version 22 software (IBM Corp., Armonk, NY, USA). To compare the amount of microleakage between enamel and dentin in each group, Mann-Whitney U test was used. The significance level was 0.05.

Results

The mean of microleakage in enamel and dentin of the four groups can be seen in Table 2. The highest amount of microleakage in enamel was for G1 and the lowest was related to G2. The highest amount of microleakage in dentin is for G1 and the lowest amount is related to G3. According to the Kruskal-Wallis test, the difference in microleakage between enamel and dentin in all groups was not significant. ($p>0.05$).

In order to compare the amount of microleakage between enamel and dentin in each group, Mann-Whitney U test was used. According to the results obtained in deciduous teeth, there is no significant difference in dye penetration between dentin and enamel (Table 3). The amount of enamel microleakage in G2 and G4 was more than this amount in dentin, while in G1 and G2, this amount is less than the dentin. However, these differences were not significant.

Discussion

Today, one of the most important factors that lead to the long-term success of restorations is to prevent microleakage between the restorative material and the surface of the enamel and dentin, but no restorative material has developed enough to completely prevent microleakage [15]. It is more important to prevent microleakage on the dentin surface, because most of the damaged teeth have a large amount of unsuitable and irreparable enamel, and in addition, the bond strength to the dentin in the conventional adhesive process compared to enamel, It is unpredictable and less, also factors such as shrinkage of the composite polymerization or contamination of the repaired surface can lead to more microleakage [16].

Antimicrobials are widely used in dental treatments such as endodontics and restoratives [17]. Antimicrobials such as chlorhexidine and desensitizers such as glutaraldehyde may also be used under composite restorations, so there is concern that the use of these materials may adversely affect the bond quality between restorations and teeth.

Numerous studies have been performed on the effect of antimicrobial and desensitizing agents on microleakage and bond strength of composite restorations. The present study is one of the studies that investigated the microleakage of composite restorations in deciduous teeth.

Owens et al. [18] in their study stated that chlorhexidine solution (Consepsis®) has a better antimicrobial effect. This solution contains 2% chlorhexidine and only its bisphenol (Contains chlorine) has stable antibacterial properties. In this study, as in many previous studies, this agent was used as an antimicrobial agent under restoration.

Also in this study, dentin desensitizer (Gluma Desensitizer®) was used, which is more effective in reducing dentin sensitivity [19]. Its main components are Glutaraldehyde and HEMA and it acts with protein deposition in dentin tubules [20].

Therefore, in this study, 2% chlorhexidine solution (Consepsis®) and glutaraldehyde solution (Gluma Desensitizer®), which are more widely used today, were used separately and at the same time, to investigate their effect on microleakage of composite restorations of deciduous teeth. If there is no negative effect on it and the results are similar or better than permanent teeth, according to the proven benefits of these materials, it is suggested to use them in pediatric treatments.

Most recent research on permanent teeth shows that the application of 2% chlorhexidine after etching on the cavity, can significantly improve microleakage [16,21]. Bin-Shuwaish et al., showed the improvement of marginal sealing on the cervical side where there is less enamel [22].

Results in bond strength also vary after the application of different antimicrobial agents. Most of the results are that the bond strength can be improved or there is no significant difference. However, researchers have suggested that more studies be done [17, 23,24].

The effect of chlorhexidine on microleakage also depends on other factors such as the technique of using acid etching. In the study of Bin-Shuwaish et al. [25], the effect of chlorhexidine for the two groups was investigated by etch-and-rinse and self-etch techniques. The result was that microleakage was significantly better in the etch-and-rinse groups than in the control group, while there was no significant difference in the self-etch group. In the present study, etch and rinse technique was used.

Contrary to the positive effects of chlorhexidine on microleakage, the use of desensitizing agents, such as glutaraldehyde, has no such effect. Microleakage was significantly increased in the study of Zarzecka-Francica et al. [20] and Daryakenari et al. [21] There was no significant difference compared to the control group. In general, microleakage studies on the use of chlorhexidine have had better results than the use of desensitizers.

These results were different in deciduous teeth. The study of Bahrololoomi et al. (6) and the study of Shafi'i et al. [26] show the amount of microleakage in chlorhexidine application increased significantly. Also in the study of Kapdan et al. [5] there was no significant difference compared to the control group, but significantly reduced the bond strength.

In this study, in addition to the application of chlorhexidine and glutaraldehyde, their combined application was also used. Only two studies were found to use these two substances together. Daryakenari et al. [21,27] n two studies on permanent teeth found the application of glutaraldehyde alone did not have a significant effect on increasing microleakage but improved the shear bond strength; in the group that used chlorhexidine alone and also chlorhexidine with glutaraldehyde, a significant reduction in microleakage was observed but there was no significant difference in the shear bond strength.

In the present study, no significant difference was observed in any of the three groups in microleakage compared to the control group ($p>0.05$). However, the mean microleakage in the chlorhexidine group was the lowest compared to the other groups and in all comparisons, the control group had the highest mean microleakage. There were also differences in enamel and dentin, so the lowest mean microleakage on enamel belonged to the glutaraldehyde group and the lowest mean microleakage on dentin belonged to the chlorhexidine group. However, these differences are not statistically significant and it seems that to achieve better and more definite results, more studies are needed with higher samples.

Also, this difference in results of deciduous teeth compared to permanent teeth can be due to the inherent and structural differences of deciduous teeth. Here are some differences between deciduous and permanent teeth that may indicate deciduous teeth are more vulnerable: the thickness of deciduous enamel is half the thickness of permanent enamel; structure of deciduous teeth is less mineralized; the enamel rods of deciduous teeth in cervical region is in occlusal direction, while in permanent teeth it is in apical direction; also pulp horns in deciduous teeth are longer and closer to DEJ and outer surface of crown [1,9].

Although in most previous studies, the negative effect of using Chlorhexidine and Glutaraldehyde solutions on microleakage or bond strength has not been observed, but for widespread use of these materials alone or in combination with restorative materials, long-term laboratory or clinical studies are required.

Bullet points

- Application of two substances (2% Chlorhexidine & Glutaraldehyde) and their combination had no significant positive effect on the microleakage of composite restorations on deciduous teeth.
- There was no negative effect of microleakage with using of two substances (2% Chlorhexidine & Glutaraldehyde) and their combination compared to the control group, so it is not a forbidden reason to using them for antimicrobial or desensitizing purposes.

References

1. Nowak AJ. (2019) Pediatric dentistry: infancy through adolescence. 6th ed: Elsevier; 2019.

2. André V. Ritter LWB, Ricardo Walter. (2019) Sturdevant's Art and Science of Operative Dentistry. 7th ed: Elsevier Health Sciences.
3. Mathew SM, Abi Mathew Thomas GK, Dua K. (2013) Evaluation of the microleakage of chlorhexidine-modified glass ionomer cement: an in vivo study. International journal of clinical pediatric dentistry. 6(1):7.
4. Hameed H, Babu BP, Sagir VM, Chiriyath KJ, Mathias J, et al. (2015) Microleakage in resin composite restoration following antimicrobial pre-treatments with 2% chlorhexidine and clearfil protect bond. J inter oral healt: JIOH. 7(7):71.
5. Kapdan A, Öztaş N. (2015) Effects of chlorhexidine and gaseous ozone on microleakage and on the bond strength of dentin bonding agents with compomer restoration on primary teeth. J Dent Sci. 10(1):46-54.
6. Bahrololoomi Z, Salajeghe S, Shahini Tiran S. (1999) Effect of Two Disinfecting Agents on the Microleakage of Resin-Based Composite Restorations in Primary Teeth. J Dent Material Tech. 8(3):107-13.
7. Yikilgan İ, Akgul S, Özcan S, Bala O, Ömürlü H. (2016) An in vitro evaluation of the effects of desensitizing agents on microleakage of Class V cavities. Journal of clinical and experimental dentistry. 8(1):e55.
8. Silveira de Araújo C, Incerti da Silva T, Ogliari F, Meireles S, Piva E, et al. (2006) Microleakage of seven adhesive systems in enamel and dentin. J Contemp Dent Pract. 7(5):26-33.
9. Dean JA. (2016) McDonald and Avery's dentistry for the child and adolescent. 10th ed: Elsevier Health Sci.
10. Teymoornezhad K, Alaghehmand H, Daryakenari G, Khafri S, Tabari M. (2016) Evaluating the microshear bond strength and microleakage of flowable composites containing zinc oxide nano-particles. Electronic physician. 8(11):3289.
11. Santini A, Ivanovic V, Ibbetson R, Milia E. (2004) Influence of marginal bevels on microleakage around Class V cavities bonded with seven self-etching agents. American journal of dentistry. 17(4):257-61.
12. Stawarczyk B, Hartmann L, Hartmann R, Roos M, Ender A, et al. (2012) Impact of Gluma Desensitizer on the tensile strength of zirconia crowns bonded to dentin: an in vitro study. Clin oral investi. 16(1):201-13.
13. Darabi F, EFTEKHARI M. (2009) Effect of chlorhexidine on microleakage of composite restorations.
14. Heintze S, Forjanic M, Cavalleri A. (2008) Microleakage of Class II restorations with different tracers-comparison with SEM quantitative analysis. J Adhesive Denti. 10(4).
15. Rathi SD, Nikhade P, Chandak M, Motwani N, Rathi C, et al. (2020) Microleakage in Composite Resin Restoration-A Review Article. J Evolution Med Dent Sci. 9(12):1006-11.
16. Motwani NM, Ikhar A, Nikhade P, Chandak M, Khatod S, et al. Dentinal Pre-Treatment in Restorative Dentistry.
17. Parisay I, Talebi M, Asadi S, Nikbakht MH. (2021) Antimicrobial Efficacy of 2.5% Sodium Hypochlorite, 2% Chlorhexidine, and 1.5% Hydrogen Peroxide on Enterococcus Faecalis in Pulpectomy of Necrotic Primary Teeth. J Dent Mater Tech. 10(2).
18. Owens BM, Lim DY, Arheart KL. (2003) The effect of antimicrobial pre-treatments on the performance of resin composite restorations. Operative denti. 28(6):716-22.
19. Mehta D, Gowda VS, Santosh A, Finger WJ, Sasaki K. (2014) Randomized controlled clinical trial on the efficacy of dentin desensitizing agents. Acta Odontologica Scandinavica. 72(8):936-41.
20. Zarzecka-Francica E, Gołda J, Górnicka O, Śliwowska B, Kustra P, et al. (2020) The influence of a selected desensitizer on marginal microleakage of a class V composite restoration subjected to thermocycles: an in vitro study. Folia Medica Cracoviensia. 43-54-43-54.
21. Sabet FÖM, Daryakenari Gh. (2021) Evaluation of the effect of chlorhexidine and glutaraldehyde on the microleakage of composite resin restorations.
22. Bin-Shuwaish M, AlHussaini A, AlHudaithy L, AlDukhiel S, AlJamhan A, et al. (2021) Effects of different antibacterial disinfectants on microleakage of bulk-fill composite bonded to different tooth structures. BMC Oral Health. 21(1):1-9.
23. Coelho A, Amaro I, Rascão B, Marcelino I, Paula A, et al. (2021) Effect of cavity disinfectants on dentin bond strength and clinical success of composite restorations—a systematic review of in vitro, in situ and clinical studies. International journal of molecular sciences. 22(1):353.
24. Hardan L, Bourgi R, Cuevas-Suárez CE, Zarow M, Kharouf N, et al. (2021) The Bond Strength and Antibacterial Activity of the Universal Dentin Bonding System: A Systematic Review and Meta-Analysis. Microorganisms.9(6):1230.

25. Bin-Shuwaish MS, AlHussaini AA, AlHudaithy LH, AlDukhiel SA, Al-Jamhan AS. (2020) An in vitro evaluation of microleakage of resin-based composites bonded to chlorhexidine-pretreated dentin by different protocols of a universal adhesive system. The Saudi Dent J.
26. Memarpour M, Shafiei F, Moradian M. (2014) Effects of disinfectant agents on microleakage in primary tooth-colored restorations: an in vitro study. J denti children. 81(2):56-62.
27. Daryakenari Gh NMN. (2021) Evaluating the Effect of %2 Chlorhexidine and %5 Glutaraldehyde Application on the Micro-shear Bond Strength of Resin Composite Restorations.