Effect of Two Disinfecting Agents on the Microleakage of Resin-Based Composite Restorations in Primary Teeth

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Received 2 February 2019 and Accepted 11 June 2019

Abstract

Introduction: Disinfection of the cavity preparation after caries excavation can lead to the elimination of bacterial remnants that can be responsible for recurrent caries, postoperative sensitivity, and failure of the restoration. The purpose of this study was to evaluate the effects of two disinfectants on microleakage of composite restorations in primary teeth. **Methods:** In this experimental study, 40 extracted primary canine teeth without caries were randomly divided into four groups of 10 teeth, namely control group (1), pretreatment group with chlorhexidine 2% (2), pretreatment group with sodium hypochlorite 2/5% (3), and pretreatment group with sodium hypochlorite 5% (4). In addition, class V cavities were prepared on all teeth. The cavities were then restored with composite. The teeth were thermocycled and immersed in 2% methylene blue for 24h. Microleakage was assessed by dye penetration in the incisal and gingival surface of the teeth using stereomicroscope. Data were analyzed in SPSS software (version 18) through Kruskal-Wallis and Mann-Whitney U tests. **Results:** According to the results, the difference among groups was significant (P<0/001). The pair-wise comparison showed significant differences between groups 1 and 2; however, no difference was observed between pretreatment groups with sodium hypochlorite 5% and 2/5%. Furthermore, no statistically significant difference was found in terms of incisal and gingival surfaces. **Conclusion:** Pretreatment with chlorhexidine and sodium hypochlorite increased microleakage in composite restorations in primary teeth.

Keywords: Chlorhexidine, Composite restorations, Disinfectant agent, Microleakage, Primary teeth, Sodium hypochlorite.

Bahrololoomi Z, Salajeghe S, Shahini Tiran S. Effect of Two Disinfecting Agents on the Microleakage of Resin-Based Composite Restorations in Primary Teeth**.** J Dent Mater Tech 2019; 8(3): 107-13.

Introduction

Today, dental caries is among the most common problems in dentistry. Moreover, it has been reported that recurrent carries are the major problems after dental restorations (1). In recent years, there has been an increase in the utilization of tooth-colored restorative materials, such as resin-based composites, glass ionomers, and the combination of both in primary teeth. Indirect pulp therapy is a method of treatment for deep carious lesions without causing degeneration of the pulp. The caries lesion is completely removed; however, the affected dentin above the pulp is retained in this treatment (2).

Research has shown that bacteria may proliferate in the smear layer and release toxins that can cause inflammation of the pulp. The topic being discussed currently is to integrate antibacterial agents into restorative material or water to eliminate the bacteria that exist in the cavity wall. Therefore, the use of antimicrobial solutions may reduce the incidence of post restorative sensitization (3). However, the effect of antibacterial agents on restorative material still raises some concern for dentists (4). Chlorhexidine gluconate is a chlorine containing bisphenol which has been used as a safe and wide spectrum disinfectant for years (3). Chlorhexidine has a broad spectrum effect on both gram-positive and gram-negative bacteria. Research has shown that chlorhexidine reduces the bacterial load in saliva and plaque as well as Streptococcus mutans levels in occlusal grooves and root surfaces (5).

Recently, it has been shown that chlorhexidine has an inhibitory effect on the endogen collagen degradation process in the dentin. Considering recent and limited in vivo and in vitro studies in this field, chlorhexidine can be used with the etching and rinse procedure as a disinfectant to enhance bond endurance. However, long term clinical studies are required to verify this (5). Sodium hypochlorite is an effective organic solution that is used vastly in clinical dentistry as a detergent. Sodium hypochlorite was first employed in the 1920s as an antimicrobial antiseptic for endodontic treatment. Sodium hypochlorite breaks down to form sodium chloride and oxygen as soon as it comes into contact with dentin which starts the oxidation process in the dentin matrix. Studies have been proven sodium hypochlorite’s antibacterial and tissue degrading effects on remnant microbes (4).

Microleakage is a dynamic process which allows the exchange of liquid, ions, molecule, debris, and microbial products across the tooth-restoration interface. Microleakage leads to post-operative sensitization of the tooth, discoloration of restoration margins, recurrent caries, and pulp damage (6). Different studies have expressed different opinions and results regarding the effect of antibacterial substances on the bonding of restorative material to dentin.

In a study conducted by Memarpour et al. (2), it was found that adding chlorhexidine to resin-based composite restorations increased microleakage.

This study was carried out to analyze the effects of antibacterial material on the microleakage resin-based composite restorations in deciduous teeth.

Materials and Methods

In this experimental study, 40 primary canines without caries were collected from patients underwent orthodontic treatment. The teeth were cleaned using non-fluoride pumice and immersed in 0.1 % Chloramine T solution for 2 weeks for disinfection. Then, the teeth were kept in distilled water during the study.

Standard class V preparations (2.5mm width, 3mm height, and 1mm depth) were performed on the buccal surface adjacent on the cementoenamel junction (CEJ) using a fissure diamond bur (008, Tizkavan Co., Iran) and high-speed handpiece with water coolant. No bevel was made in the cavity preparation. The incisal margin was placed on enamel and the gingival margin on cementum.

The teeth were then randomly allocated into four groups of 10 teeth, and each group was specified with a certain antibacterial agent. Group 1 (control group) was etched with 37% phosphoric acid (Diadent, Korea) for 15 seconds and then rinsed for 15 seconds and dried with a low-pressure air syringe. Two layers of the etch and rinse Adper single bond 2 (3M, ESPE, USA) as well as adhesive were applied and thinned with low-pressure air syringe cured for 20 sec with a LED light cure system (Woodpecker, China) with an intensity of 600 mW/cm^2. The teeth were restored using resin-based composite (Z250, 3M, ESPE, USA) and cured for 40 seconds. The light intensity was measured using a light intensity meter.

The cavities in group 2 were etched, rinsed, and dried similar to the former group. Then, a thin layer of 2% non-alcoholic chlorhexidine (Maquira, Brazil) was applied for 60 seconds. After drying with air for 10 seconds, the cavity was restored after bonding using resin-based composite, similar to group 1.

Similar cavity preparation was applied to group 3 and the cavities were etched, rinsed, and dried, subsequently, a thin layer of 2.5 % sodium hypochlorite was applied for 15 seconds and then dried with air. After bonding was applied, the cavity was restored using resin-based composite in a similar procedure to that in previous groups.

Group 4 was subjected to similar steps used in previous groups. A thin layer of 5.25% sodium hypochlorite was applied after etching, rinsing, and drying. After drying with air for 10 seconds, the teeth were restored using the same procedure applied to previous groups. All teeth were stored in distilled water at room temperature and thermocycled (Vafayi, Iran) 5°C and 55°C for 5000 cycles with a 30-second dwell timethermocycling.(After thermocycling, the samples from each group were prepared to be placed in staining solutions as follows:

The apexes of the teeth were sealed with sticky wax, and the root and crown surfaces were covered with 2 layers of nail polish at 1 mm margin of the cavity to prevent microleakage from other areas and interfering with the area under study. All the teeth were then separately immersed in 2% methylene blue solution for staining and kept at room temperature for 24 h. The samples were rinsed and cut faciolingually from the middle of the restoration using a diamond disc and water coolant (2). Two examiners calibrated with each other examined the linear penetration of the stain under a microscope (ZTX-3E, China) with a magnification of 20x. In order to increase the accuracy of the results, both segments of each tooth were examined, and the highest value was reported as the microleakage score.

The scoring system for microleakage is as follows:

Score 0: No penetration

Score 1: Penetration up to 1/3 of the cavity depth

Score 2: Penetration up to 2/3 of the cavity depth

Score 3: Complete penetration of the cavity depth but not the axial wall

Score 4: Penetration of stain into the axial wall.

Data were analyzed in SPSS software (version 18) through Kruskal-Wallis test for all groups and Mann-Whitney U test for paired difference test with a pre-set significance level of 0.05.

Results

This study was conducted on 40 extracted primary canines without caries which were divided into four groups.

Each sample was examined regarding incisal and gingival surfaces by two observers. Statistical analysis showed that there was no statistically significant difference between the two examiners (P>0.05).

The normality of the data was tested using Shapiro-Wilk test and the results showed the non-normal distribution of data in the test groups and both cervical and incisal surfaces (P<0.001).Therefore, non-parametric tests were used in this study.

Comparison of the mean values of composite microleakage showed a statistically significant difference among the four groups in terms of both gingival and incisal surfaces (Tables I, II). The highest and lowest levels of microleakage were observed in 2.5% sodium hypochlorite and the control group, respectively, regarding both gingival and incisal surfaces. Furthermore, there was no significant difference between the incisal and gingival levels regarding the mean of microleakage in any of the groups (Table III). The total mean values of microleakage in the gingival and incisal surfaces were 2.60±1.61 and 2.40±1.90, respectively. In addition, no statistically significant difference was observed regarding the comparison of mean values of microleakage in these surfaces (P=0.715).

Table IV summarizes the results obtained from the paired difference tests. According to the results, there was a significant difference between the control group and the other three groups regarding chlorhexidine, 2.5% sodium hypochlorite, and 5% sodium hypochlorite in both gingival and incisal surfaces. Consequently, the microleakage of all three groups was higher than that in the control group. However, there was no significant difference among the three groups in terms of the abovementioned variables.

Table I. The mean values of microleakage in four groups regarding incisal surface

Microleakage value Group	Number of samples	Mean	Standard deviation	Minimum	Maximum	P-value
2% chlorhexidine	10	2.70	1.89	0	4	
5% sodium hypochlorite	10	2.60	1.84	0	4	
2.5% sodium hypochlorite	10	3.60	1.26	0	4	
Control	10	0.70	1.49	0	4	0.004
Total	40	2.40	1.90	0	4	

Table II. The mean values of microleakage in four groups regarding gingival surface

Microleakage value Group	Number of samples	Mean	Standard deviation	Minimum	Maximum	P-value
2% chlorhexidine	10	3.10	1.66	0	4	
5% sodium hypochlorite	10	3.00	1.25	0	4	
2.5% sodium hypochlorite	10	3.20	1.69	0	4	
Control	10	1.10	0.88	0	3	0.007
Total	40	2.60	1.61	0	4	

Table III. Comparison of the mean microleakage in four groups regarding the incisal and gingival levels

Microleakage value	Incisal		Gingival		P-value
Group	Mean	Standard deviation	Mean	Standard deviation	
2%chlorhexidine	2.7	1.89	3.1	1.66	0.622
5%sodium hypochlorite	2.6	1.84	3	1.25	0.968
2.5%sodium hypochlorite	3.6	1.26	3.2	1.69	0.542
Control	0.7	1.49	1.1	0.88	0.065
Total	2.4	1.90	2.6	1.61	0.715

Table IV. Paired difference test of groups with each other in terms of microleakage value

First group	Second group	P-value (Incisal)	P-value (Gingival)
Control	Chlorhexidine	0.019	0.013
	2.5% Sodium hypochlorite	0.001	0.011
	5% Sodium hypochlorite	0.026	0.004
Chlorhexidine	2.5% Sodium hypochlorite	0.147	0.689
	5% Sodium hypochlorite	0.768	0.357
2.5% sodium hypochlorite	5% Sodium hypochlorite	0.075	0.198

Discussion

This study investigated the microleakage of resin-based composite restorations in deciduous teeth using two types of antibacterial agents, namely chlorhexidine and sodium hypochlorite. The microleakage was tested utilizing the dye penetration method. Dye penetration is the most common method for this purpose which has advantages, such as low cost and easy methodology. However, and disadvantages include subjective analysis of results and the low molecular weight of the dye which is less than that of the bacteria. This drives the dye to penetrate into areas which bacteria cannot (1).

There is controversy regarding the application of chlorhexidine prior to or after etching. In a study conducted by Vieira et al. (7), chlorhexidine was used prior to etching. However, Tulunopglu et al. (8) and Ersin et al. (9) applied chlorhexidine after etching which was similar to this study.

The reason for this was to increase the antibacterial agent's effect by applying after the removal of the smear layer and exposure of the underlying tissue. The use of chlorhexidine after etching and before applying the adhesive system can lead to the inhibition of matrix metalloproteinase as well as reducing the bacterial load.

However, De Castro et al. (10) and Soares et al. (11) concluded in their studies that the use of 2% chlorhexidine before or after etching had no statistically significant effect on tensile bond strength of dentin in permanent dentition.

Breschi et al. (12) reported that applying 0.2% chlorhexidine was as effective as 2% chlorhexidine in maintaining bond strength in permanent teeth after a 12-month analysis. Furthermore, Manfro et al. (13) suggested that low concentrations of chlorhexidine can be effective in slowing down the debonding that occurs at the adhesive interface. Previous studies have claimed that the use of 0.12% to 0.2% chlorhexidine before or after etching does not have detrimental effects on immediate bond strength in primary and permanent teeth (7).

According to the results of these studies, 2% chlorhexidine concentration was used in this study. Throughout different studies, it has been shown that the application of chlorhexidine for one minute to the etched dentin after rinsing the acid etch in the total etch bonding system can be an effective technique in increasing the longevity of the dentin-resin bond(12). Therefore, in this study, chlorhexidine was placed in contact with dentine for 60 seconds to induce its effect. Independent of the volume and duration that chlorhexidine is applied to, a considerable amount remains in the dentin matrix (14).

In the current study, 40 primary canines without caries were restored and examined at the gingival and incisal surfaces. The median microleakage score was 4 in 2% chlorhexidine and 2.5% sodium hypochlorite groups which were the maximum score for penetration. In a 5% sodium hypochlorite group, the median score was 3 which shows high penetration and leakage.

However, in the control group, the median score was obtained at 0.5 which showed the lowest level of microleakage. In this study, both disinfecting agents increase the microleakage and there was no statistically significant difference between the scores of chlorhexidine and both percentages of sodium hypochlorite. The undesirable effects of chlorhexidine in terms of sealing the etched dentin may be due to the interference of remaining chlorhexidine with the mineral content of dentin, such as calcium, and phosphate (2).

The results of a study performed by Memarpour et al. (2) correlated with the findings obtained from this study. Memarpour et al. (2) used deciduous teeth; however, the antibacterial agent was chlorhexidine, and Clearfil protect bond was used in the abovementioned study. Besides microleakage, the effect of antibacterial agents on glass ionomer was also analyzed in this study (2).

The results of the current study contradicted those of several other studies (3, 13, 15, 16). Nevertheless, in a study by Kapdan et al. (3), compomer restorations in primary teeth were analyzed, and the antibacterial agents were chlorhexidine and ozone gas. The results showed no statistically significant difference among the groups in terms of microleakage.

The other major difference in this study was the use of self-etch bonding system (prime&- bond NT). Ricci et al. (16) reported in a clinical study that chlorhexidine had no negative effects on the bond of resin-based composite restorations in primary teeth in the initial months after the restorative procedure. Moreover, Manfro et al. (13) stated that using different percentages of chlorhexidine did not significantly affect the bond strength of adhesive to dentin in primary teeth. The obtained results are not consistent with the findings of the present study. Even though, the test method (bond strength) is different to that of our study (17).

Considering the results of a study performed by Leitune et al. (18), it can be said that chlorhexidine application after etching and before bonding increased bond strength after 6 months in primary teeth; however, the results are not in line with the findings in this study. In the same line, Sung et al. (19) reported no effect on microleakage in a study that used chlorhexidine as a rinsing solution. Additionally, in a study carried out by Ramezanian et al. (14), ethanol was used with chlorhexidine in permanent teeth. According to the results, ethanol with chlorhexidine had the potential to reduce microleakage in the long term.

The only study that was similar to the current one was a study conducted by Salama et al. (20) which was carried out on the analysis of sodium hypochlorite and chlorhexidine and primary teeth as well as the effect of Persica on microleakage of resin-based composite restorations. The results were contradictory to the findings of the present study since there was no significant difference among the microleakage of three groups, and cervical margins showed higher microleakage than incisal margins. The samples used in this study consisted of mild to moderate caries which affected microleakage scores. In addition, the concentration of used sodium hypochlorite was 1.3 % (20).

In this study, there was no statistically significant difference between incisal and gingival surfaces regarding the mean microleakage score which correlated with the results obtained from a study by Kapdan et al. (3). This discrepancy can be attributed to the fact that Kapdan (3) analyzed the effect of ozone and chlorhexidine on bond strength and microleakage. Trowbridge et al. (21) claims that the location of the lesion on the tooth is also important and affects microleakage scores since the CEJ is more permeable than enamel and dye penetrations occur more easily. To summarize, the differences in results can be explained by diversities in adhesives, analysis tests, antibacterial agents, concentrations, and the experience of the operator. One of the limitations of this study was the lack of aging processes on the samples and it is suggested that further studies be carried out utilizing more concentrations and antibacterial agents.

Conclusion

The utilization of antibacterial agents, such as chlorhexidine and sodium hypochlorite in resin-based composite restorations in deciduous teeth leads to an increase in microleakage.

Conflict of Interest

There is no conflict of interests regarding the publication of the article

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