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ORIGINAL RESEARCH

Evaluation of the Influence of Three different Temperatures on Microleakage of Two Self-etch and One Total-etch Adhesives

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ABSTRACT

Aim: To evaluate the bonding temperature effect on dentin-restoration microleakage.

The null hypothesis of the study is that the score of microleakage is identical among different adhesive bondings at different temperatures.

Materials and methods: Ninety caries free maxillary premolars were selected. Class V cavities were prepared on the cemento enamel junction (CEJ) of the buccal sides with enamel margins on occlusal sides and cementum margins on gingival sides. The specimens were divided into 3 groups: G1, single bond adhesive + Z250 composite; G2, P90 adhesive + Filtek Silorane composite; and G3, Clearfil SE bond + Clearfil APX. All groups were divided into three subgroups based on the adhesive temperature: A—4°C; B—25°C; and C—40°C. After coating the specimens with nail polish 1 mm beyond the margin of the restorations, they were stored in 0.5% basic Fuchsin dye solution for 24 hours. The teeth then were buccolingually sectioned and observed under a stereomicroscope.

Results: There was no significant difference between microleakage of occlusal and gingival margins in each group. Clearfil SE bond and Adper single bond displayed lower microleakage than P90 adhesive at 4°C and 25°C. The most and least microleakage score for Adper single bond was at 40°C and 25°C respectively. Clearfil SE bond showed less microleakage at 25°C than 4°C and 40°C.

Conclusion: Clearfil SE bond and Adper single bond displayed less microleakage at 25°C while there was no significant difference among for P90 adhesive microleakage at three temperatures.

Keywords: Adhesive temperature, Microleakage, Silorane composite.

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INTRODUCTION

Intimate contact between composite and dentin is critical for clinical success of composite restoration. Microleakage between tooth and composite causes diffusion of fluids and microorganisms through it, which can lead to restoration failure.[1-3]

Self-etch adhesives, are classified as two-step and one-step systems. These kinds of adhesives have more complex formulations, and are more users friendly as well.[4] As the self-etch monomers have less acidity for dissolving dental minerals than phosphoric acid, they leave more minerals attached to dentin collagen fibers which can cause additional chemical adhesion between dentin and adhesive monomers and less microleakage as a result. There are different types of self-etch adhesives according to their pH value: Ultramild (pH $\approx$ 2.5), mild (pH $\approx$ 2), intermediary strong (pH $\approx$ 1.5), and strong (pH $\leq$ 1).[5] There was reported that mild self-etch adhesives shows lower nanoleakage than strong ones.[6]

Due to low viscosity, adhesive agents have more wettability and penetration in dental substrate.[7] The vapor of adhesive solvent depends on temperature.[8] Increasing the temperature might increase more solvent evaporation and decrease the monomers viscosity.[9,10] It was reported that low temperature can decrease solvent evaporation. Besides, it increases polymeric material viscosity.[4]

Several studies evaluated resin composite temperature effect on bond strength before using on dental substrate. They showed that prewarming to 55 to 60ºC could increase monomer conversion.[11,12] The effect of temperature on some adhesive agents has been studied as well.[13,14] Some studies showed higher bond strength of self-etch adhesives was in cold temperature while others reported that the highest bond strength for these adhesives was in heated temperature.[4,14,15] Since the bond strength value does not necessarily correlate with marginal adaptation and there are not specific results reported for microleakge of adhesives in different temperature, a microleakage study seems to be necessary.[16]

Silorane composites are recently introduced for clinical use. Their bonding agent is a two bottle self-etch adhesive.[17] Some studies showed higher marginal adaptation and lower cuspal deflection for silorane composites.[18] Microleakage of Filtek Silorane system (pH = 2.7) was reported significantly lower than Prompt L Pop/Z250 (pH = 0.8) which may be the result of adhesive chemical adhesion to dentin minerals due to lower pH of P90 adhesive.[19] In addition, the low shrinkage characteristics of Filtek Silorane composite which causes less gap formation in the dentin-composite interface may cause lower microleakage of P90. However, the effect of temperature on adhesive Siloran microleakage is not reported yet.

Storage temperature can influence on self-etch adhesives hydrolitic stability more than total-etch adhesives. It is reported that cold temperature shows more shelf-life in self-etch adhesives.[20,21] So adhesive bondings especially self-etch adhesives might be stored in refrigerator in order to extend their shelf-life; adversely, they might be kept at room temperatures or warm areas.[13]

pH difference in self-etch adhesives may affect their microleakage in cold versus heated temperature. The aim of this study is to evaluate the different temperatures on microleakage of composite restorations with two different self-etch adhesives (P90 Silorane adhesive and Clearfil SE bond) and one total-etch adhesive (Adper single bond). The null hypothesis of the study is that the score of microleakage is identical among different adhesive bondings at different temperatures.

MATERIALS AND METHODS

Ninety caries-free extracted human maxillary premolars were selected and initially stored in saline solution and immersed in a 0.1% thymol solution for 24 hours prior to the onset of the experiment. Then they were mounted in acrylic resin (3 cm width, 5 cm height) 3 mm above the CEJ. Class V cavity was prepared (1.5 mm depth, 2 mm occlusal-gingival, 3 mm mesio-distal) using fissure bur on the CEJ of buccal side of each tooth with the occlusal margin on enamel and the gingival margin 1 mm below the CEJ. Adhesives and resin composites used in this study are presented in Table 1.

Teeth were randomly divided into 3 groups (N = 30); group 1: single bond + Z250 (3M ESPE/USA), group 2: P90 + Filtek Silorane (3M ESPE/USA), and group 3: Clearfil SE Bond + Clearfil AP-X (Kuraray, Japan). All the groups were further divided into 3 subgroups (N = 10) based on the bonding temperature; Subgroup A: 4°C, subgroup B: 25°C, and subgroup C: 40°C.

The primer and adhesive bottles in the subgroup A, B, and C were put in refrigerator at 4°C for 30 minutes, room temperature (25°C) for 15 minutes, and in warm bath water (TE04 mag, Technal, Piracicaba, Brazil) at 40°C for 15 minutes respectively.[22] The adhesives were applied following the manufactures instructions (Table 1). After applying adhesive agents in each cavity, composite resins was inserted and cured for 40 seconds with LED curing unit 1500 mw/cm^2 (Kerr, Demi).

Then, after 24 hours storage of specimen in distilled water at room temperature they were thermocycled for 1000 cycles in thermocycler (Vafaee, Iran) between 5 to 55ºC with 30 seconds dwell time and 30 seconds interval time. Then teeth were coated with two layers of nail polish except 1 mm around the restoration. They were immersed in 0.5% basic fuchsin dye solution for 24 hours. After washing the teeth with tap water, the root was removed by sectioning at the border of acrylic mount (3 mm under the CEJ) and sectioned buccolingually at the mid of restoration with saw and water coolant (leitz 1000, Wetzlar, Germany). The sections were observed

Table 1: Materials used in the study

Materials	*Composition*	*Application mode*
Filtek Silorane (3M ESPE, USA)	Primer: Phosphorylated methacrylates, vitrebond copolymer, BisGMA, HEMA, water, ethanol, Silorane-treated silica filler	applied for 10 seconds, air blowed, polymerized for 10 seconds
	Adhesive: Hydrophobicdimethacrylate, phosphorylated methacrylates, TEGDMA, Silorane-treated silica filler	
Clearfil SE bond (Kuraray, Japan)	Primer: MDP, HEMA, hydrophilic dimethacrylate, Water dl-camphorquinone, n, n-diethanol-p-toluidine	Applied with a brush, gently air dried.
	Bond: MDP, bis-GMA, HEMA, hydrophobic dimethacrylate, dl-champhorquinone, N, N-diethanol p-t oluidine, silanated colloidal silica	Bonding agent applied, gently air dried, light cured for 10 seconds
Adper single bond (3M ESPE, USA)	Bis-GMA, HEMA, Copolymer of acrylic and itaconic acids, glycerol 1, 3-dimethacrylate, 5-1 Diurethane dimethacrylate Silane treated silica, Ethyl alcohol, Water	Acid etching, bonding agent applied for 10 seconds in 2 layers and light cured for 20 seconds
Filtek Silorane composite (3M ESPE, USA)	Bis-3, 4-Epoxycyclohexylethylphenylmethylsilane, 3,4 Epoxycyclohexylcyclopolymethylsiloxane, silanized, quartz	Apply resin composite in 2 mm layers, cure for 40 seconds
Clearfil AP-X (Kuraray, Japan)	Bis-GMA, TEGDMA, Barium glass, silica	Apply resin composite in 2 mm layers, cure for 20 seconds
Filtek Z250 (3M ESPE, USA)	Bis-GMA, UDMA, Bis-EMA, TEGDMA, zirconia, silica	Apply resin composite in 2 mm layers, cure for 20 seconds

Table 2: Microleakage scores in gingival and incisal margins

Margins	*Gingival*					*Occlusal*				
Groups / *Scores*	*0*	*1*	*2*	*3*	*Mean (SD)*	*0*	*1*	*2*	*3*	*Mean (SD)*
Single bond 4°C	3	2	10	0	51.83	2	11	2	0	33.77
Single bond 25°C	11	3	1	0	23.77	12	3	0	0	13.70
Single bond 40°C	0	0	5	10	97.17	1	0	8	6	88.97
P90 4°C	1	0	11	3	71.73	0	0	3	12	100.70
P90 25°C	1	0	1	13	103.40	0	4	9	2	62.57
P90 40°C	1	0	0	14	106.57	0	2	0	13	99.23
Clearfil SE bond 4°C	3	11	1	0	58.87	0	1	5	9	94.83
Clearfil SE bond 25°C	11	3	0	1	26.93	3	8	3	1	35.13
Clearfil SE bond 40°C	1	0	11	3	71.73	0	0	6	9	83.10

SD: Standard deviation

under a stereomicroscope at 40 magnifications (oral Zeiss 2 nc, Oberkochen, Germany) by two observers separately.

The dye penetration recorded based on this scoring system;[23]

0 = no dye penetration
1 = dye penetration involving half of occlusal/ gingival wall
2 = more than half of occlusal/ gingival wall
3 = penetration along the axial wall.

Then statistical analysis was done using Mann-Whitney and Kruskal-Wallis tests at $p < 0.05$.

RESULTS

The mean microleakage score of all groups are listed in Table 2. No significant difference was shown between microleakage of occlusal and gingival margins of restorations in each group according to the Mann-Whitney test. ($p < 0.05$) Kruskal-Wallis test was used for comparison between subgroups. At both 4ºC and 25ºC no significant difference was shown between Clearfil SE bond and Adper single bond ($p < 0.05$), while both of these adhesives displayed lower microleakage than P90 bond in occlusal and gingival margins at these temperatures ($p > 0.05$).

In Adper single bond subgroup there was a significant difference between microleakage of gingival and occlusal margins at three temperatures (25ºC < 4ºC < 40ºC) $p > 0.05$. In Clearfil SE Bond subgroup, there was significantly lower microleakage at 25ºC than 4ºC and 40ºC ($p > 0.05$), but the results were not statistically different between 4ºC and 40ºC ($p < 0.05$). In P90 bonding no significant difference was shown among the three temperatures ($p < 0.05$).

DISCUSSION

Inadequate sealing of restorations causes recurrent caries, marginal staining, and postoperative sensitivity in tooth colored restorations.[2] There are many factors that influence the restoration bonding. One of them which can be evaluated more is the pre-work temperature of adhesive bonding. Some studies have reported clinical factors, such as environmental temperature and humidity can influence on early bond strength to dentine.[24,25] The three storage temperatures in this study were selected among office environments and previous studies.[4]

The adhesive of Silorane Filtek system is a two-step self-etch methacrylate based system containing a self-etching primer and a hydrophobe adhesive resin. P90 primer (pH = 2.7) causes mild etching of tooth and bonds chemically to the hydroxyapatite crystals.[17] Silorane composites have ring opening polymerization and most favorable shrinkage characteristics which causes best cavity adaptation.[26] Although it is expected that Silorane Filtek group shows lower microleakage scores than two other groups, microleakage of P90 adhesive was more than Adper single bond and Clearfil SE bond at all temperatures and the preheating temperatures did not show significantly different results in P90 microleakage in our study. The P90 primer and bondings are photoactivated as separate layers, unlike the other two-step adhesive used in this study (Clearfil SE bond) which can affect the bond durability.[26] Besides, the higher PH value of P90 primer might cause less smear layer removal and less penetration of adhesive which can cause higher microleakage than Clearfil SE bond.

Donmez et al[14] reported that microtensile bond strength of SE bond stored at 40ºC was significantly lower than 23ºC and 4ºC, while these two recent temperatures did not show different results. In their studies the storage time of adhesive was 1 year in these three temperatures, which can affect the results. In another study on microtensile bond strength of different adhesives to enamel at 5ºC, 20ºC, 40ºC, Clearfil SE bond showed higher bond strength than prime and bond NT and Adper prompt L pop at cold temperature. While prime and bond NT showed highest bond values in heated group.[4] Authors explained this by the bonding pattern of self-etch adhesives which means higher temperature impairs dissolution of smear layer by clearfil SE bond. In addition, etch and rinse bondings show increase of wettability by increasing the temperature.[27] Since the microtensile bond strength results did not show correlation with microleakage, the results of these studies and our study are not the same.[16] In microtensile bond strength studies higher temperature usually caused more bond strength for etch and rinse adhesives, for instance Prime and Bond 2.1 (acetone based) and single bond (ethanol based) showed highest early microtensile bond strength at 37ºC and 50ºC respectively; while self-etch adhesives showed higher microtensile bond strength at cold temperature.[4,15] Although in our microleakage study the etch and rinse adhesive showed the lowest and highest microleakage at 25ºC and 40ºC respectively, while P90 self-etch adhesive microleakage was similar at three temperatures and lowest microleakage scores for Clearfi SE bond were detected at 25ºC. So, increasing the temperature might result in increase of both microleakage and microtensile bond strength of etch and rinse adhesives. It is reported that self-etch adhesives showed higher bond strength at cold temperature than heated ones; however, in our study microleakage of self-etch adhesive Clearfil SE bond was not statistically different from etch and rinse adhesive but was lower than the other self-etch adhesive (P90 Silorane) at cold temperature.

Self-etching primers usually contain acidic monomer (MDP), HEMA and water. Clearfil SE bonding is composed of 25 to 30% MDP (monoester group of acid phosphoric) with pH = 2. One study reported that HEMA is hydrolyzed to methacrylic acid and ethylene glycol in acidic conditions, which is influenced by high storage temperature.[28] Higher microleakage in Clearfil SE bond at 40ºC may be a result of this chemical reaction. Chemical deterioration of adhesives (HEMA, MDP, BIS, GMA, DMA) can effect on the adhesive durability and decrease their bond strength as well.[29,21] Clearfil SE primer has low viscosity monomers and high hydrophilic concentration. In one study, Clearfil SE bond showed lower microleakage than another self-etch adhesives with pH around 1.5 which can be related to the chemical interaction with the remaining hydroxyapatite in Clearfil SE bond mild selfetch adhesive.[30] It seems that lower microleakage of Clearfil SE bond at 25ºC than P90 bonding in our study is related to this chemical interaction between Clearfil SE bond and dental hydroxyapatite residues.

A study which evaluated bond strength of self-etch and total-etch adhesives on bur prepared and smear layer free showed that etch and rinse adhesive and strong self-etch adhesive bond strength between bur prepared and smear layer free dentin was not statistically different, but both ultra mild and mild self-etch adhesives revealed lower bond strength to bur prepared dentin than smear layer free dentin.[31] All specimen in our study were prepared with bur, while self-etch ultra mild adhesive showed higher microleakage than etch and rinse group and mild self-etch adhesive which can be the result of smear layer presence on enamel and dentin substrate.

Adhesives have the ability of volatilization and penetration due to their low viscosity. Low temperature can cause a reduction in solvent evaporation which leads to lower bond strength because of porosities that form inside the adhesive layer. In addition, a thicker adhesive layer could be formed in lower temperatures of adhesives.[13,32] In one study, thicker adhesive layer showed more stress absorption and less marginal leakage.[33] Although Adper single bond and Clearfil SE Bond showed higher microleakage at 4ºC than 25ºC in our study. Choi and others used multiple adhesive layers for creating a thick adhesive layer beneath composite restoration, while we used only one layer of adhesives on cavity surfaces which does not form such a thick adhesive layer even at cold temperature applied in our study.[33]

Spreading velocities of etch and rinse adhesives increase with increasing the temperature which can cause deeper penetration rate into acid-etched dentin. In one study, increasing single bond temperature from 25ºC to 37ºC, increased the spreading velocity of it about 21%.[27] In our study Adper single bond showed higher microleakage at 40ºC than two other temperatures. This may be due to lack of a direct correlation between microleakage score and spreading velocity of adhesive.

The solvents of adhesives are different (ethanol, water or acetone). Each of these solvents has different boiling temperatures. In this study, SB and P90 primer have ethanol/water as solvents, while the solvent of SE bond is ethanol.[4,10] It seems that similar solvents couldn't affect the results of microleakage of tested groups at different temperature.

In one study, etch-and-rinse system showed higher marginal seal than self-etch adhesive at both enamel and dentin margins.[34] In our study, Adper single bond showed better occlusal and gingival seal than P90 adhesive in room and cold temperature, but the results were not statistically different from Clearfil SE bond. This is in agreement with some studies that concluded Clearfil SE Bond microleakage in enamel and dentin margins is similar to total-etch adhesives.[35,36]

As the self-etch adhesives in this study have nearly the same solvents and are classified as mild and ultramild adhesives, it is suggested to evaluate the use of self-etch adhesives with strong acidity and acetone solvent in future studies as well.

CONCLUSION

Different temperatures did not show significant difference in P90 Silorane adhesive system, while Clearfil SE bond and Adper single bond showed better adaptation at 25ºC than two other temperatures in decreasing microleakage. P90 adhesive system showed more microleakage at all temperatures than two other adhesives. According to the results, the best temperature for using adhesives is the room temperature (25ºC).

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