

Original Article

The effect of sodium hypochlorite enamel pretreatment on the shear bond strength of fissure sealant using a resin-modified glass ionomer cement and a fluoride-releasing self-etch resin adhesive

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ABSTRACT

Background: The relative high caries risk of pits and fissures highlights the importance of protecting these areas. The aim is to determine the effect of sodium hypochlorite (NaOCl) on the shear bond strength (SBS) of resin-based pit and fissure sealant (RBPFS) material to enamel using resin-modified glass ionomer cement (RMGIC) and fluoride-releasing self-etch resin (FRSE) adhesives.

Materials and Methods: In this *in vitro* study extracted third molar teeth without carious lesions or defects were divided into five experimental groups ($n = 20$). Group A: (Control group) etch (35% phosphoric acid) for 15 s and RBPFS applied. Group B: Etch for 15 s, FRSE and RBPFS applied. Group C: Pretreated with 5% NaOCl and similar steps to Group B. Group D: Etch for 15 s then RMGIC bonding agent and RBPFS applied. Group E: Pretreated with 5% NaOCl and then similar steps to Group D. SBS was determined using a universal testing machine. The tested specimens were examined under a field-emission scanning electron microscope. Data were analyzed using one-way ANOVA and post hoc Tukey's tests ($P=0.05$).

Results: A statistically significant difference between the test groups was observed; Group C showed the highest SBS mean value (7.52 ± 2.74 MPa) and Group D showed the lowest (4.48 ± 1.81 MPa) ($P < 0.001$). Pretreatment with NaOCl increased the SBS of fissure sealant when Riva bond LC was used ($P = 0.049$).

Conclusion: The use of NaOCl as pretreatment can increase the SBS of RBPFS to enamel using RMGIC adhesive. FRSE adhesive did not show improvement in SBS values using pretreatment.

Key Words: Pit and fissure sealants, pretreatment, shear strength, sodium hypochlorite

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INTRODUCTION

Cariou lesions in the pits and fissures account for approximately 80% of total caries experience.^[1] The high prevalence of caries in these areas is primarily due to pit and fissure morphology, making them difficult

to clean and also less exposed to salivary clearance, thereby increasing caries susceptibility compared to more easily cleansed smooth surfaces.^[2] Treatment

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of carious lesions, especially in children, requires advanced clinical skills and sometimes involves the high cost of sedation techniques or general anesthesia for patient management.^[3] The placement of pit and fissure sealants (PFS) is a useful measure to prevent carious lesions in these susceptible areas.

PFS application involves the placement of material into the pits and fissures, providing a physical barrier that makes the area cleansable and isolates microorganisms at the base of the fissure from the cariogenic substrate.^[4] Resin-based PFS (RBPFS) prevent carious lesion formation in clinical trials.^[5-8] The retention and therefore, the effectiveness of RBPFS is directly related to the strength of the micro-mechanical bond between the sealant material and the enamel. Higher shear bond strength (SBS) is associated with increased retention and more effective prevention.^[9] To improve the retention and quality of RBPFS, use of an intermediate bonding layer has been advocated.^[10] The dehydration activity of solvents in dental adhesives displaces water from deep fissure walls and allows deeper penetration of adhesive and sealant.^[11] In this regard, use of a fluoride-releasing adhesive is a novel approach as these materials may decrease the incidence of carious lesions associated with fissure sealants.^[12]

Clearfil Protect Bond™ (CPB; Kuraray, Osaka, Japan), is a fluoride-releasing self-etch resin (FRSE) bonding agent, that combines the physical advantages of dental adhesive technology and caries preventive effects.^[13] Riva Bond LC™ (RBLC; SDI, Vic, Australia) is a resin-modified glass ionomer cement (RMGIC)-based adhesive that has fluoride-releasing potential. Based on the manufacturer's claim, the continuous release of fluoride and the ability to compensate for the volumetric polymerization shrinkage of resin-based restorative materials make this material a potentially useful adhesive in numerous situations.^[14]

Due to the importance of RBPFS retention for caries prevention, other nondestructive techniques along with the adhesive application can be used to improve adhesion/retention. Pretreatment of the enamel surface with sodium hypochlorite (NaOCl) has been reported to increase the degree of penetration of adhesives and sealants into the enamel.^[15,16] Deproteinization of enamel surface using 5.25% NaOCl for 1 min before acid etching is effective in increasing the SBS of resin to the enamel by improving the etch pattern and bond strength.^[17-21]

However, there are limited data about the effect of NaOCl pretreatment on SBS of fissure sealants in conjunction with fluoride releasing adhesives as intermediate bonding agents. The present study aimed to evaluate the effect of NaOCl pretreatment on SBS of fissure sealant to enamel using RMGIC and FRSE adhesives.

MATERIALS AND METHODS

One hundred extracted third molar were selected from an existing pool of extracted human teeth in the Biomaterial Research Center, Shiraz, Iran for this original *in vitro* study. Ethical approval (S.66) was obtained from the Ethics Committee of the Shiraz Dental School, Iran. The selected teeth were inspected visually when wet and confirmed as teeth with enamel free of discoloration, carious lesions, and developmental defects. The roots were removed perpendicular to the long axis of the tooth with a 0.3-mm diamond blade (Minitom, Struers A/S, Copenhagen, Denmark).

The crowns were sectioned in a mesiodistal direction using the same blade, with more than one specimen obtained from an individual tooth. Specimens were manually wet polished with a circular motion in a flat surface using 600 grit silicon carbide paper (Struers A/S, Copenhagen, Denmark) to produce a flat enamel surface with as little enamel removal as possible. The prepared flat surface was placed in contact with a glass slide and stabilized with sticky wax. A plastic ring of 15-mm internal diameter was placed over the specimen and filled with self-curing acrylic resin (Acropars, Marlic Co., Tehran, Iran). Once set, the glass slide was removed, creating a supported flat enamel surface suitable for bonding. The teeth were stored in 1% chloramine-T (Sigma-Aldrich Co., MO, USA) for 24 h before initiating the experiments. Enamel specimens were divided into five experimental groups ($n = 20$).

Group A: control group (no bonding [NB]): 35% phosphoric acid etch for 15 s, rinsed for 15 s and dried with oil-free triplex air. Then, RBPFS (Clinpro Sealant; 3M Espe, MN, USA) was applied in a clear Teflon cylinder (2.65 mm in diameter and 3 mm in length) without using a bonding agent.

Group B (CPB): After etching with 35% phosphoric acid, fluoride-releasing resin-based adhesive CPB was applied according to the manufacturer's instructions. Then, RBPFS was applied. Initially, the primer was

applied, left for 20 s, air-dried and then, adhesive was applied and light cured.

Group C (CPB + NaOCl): 5.25% NaOCl (manufacturer's details) applied with a micro-brush with back and forth rubbing motion for 1 min; then, CPB was applied with similar steps to Group B.

Group D (RBLC): Enamel surface had Riva conditioner (37% Phosphoric acid for 5 s) applied; then capsules of bonding agent were activated and mixed using an ultimate 2 amalgamator (SDI, Vic, Australia) for 10 s. RBLC was applied by micro-brush and cured for 20 s; then, RBPFS was applied.

Group E (RBLC + NaOCl): 5.25% NaOCl applied with micro-brush with back and forth rubbing motion for 1 min. RBLC was applied with similar steps to Group D; then RBPFS was applied.

In all five groups, a clear Teflon™ cylinder measuring 2.65 mm in diameter and 3 mm in length, was secured to the lapped tooth surface and served as a mold into which the RBPFS was inserted.

The RBPFS was cured for 20 s from each angle according to the manufacturer's instruction. The specimens were stored in distilled deionized water for 48 h at 37°C and then SBS values were determined with a universal testing machine (Zwick/Roll Z20, Zwick GmbH and Co, Ulm-Eisingen, Germany) The test was performed by securing the specimens in a mounting jig and a sharp straight-edge chisel attached to the cross-head was used to apply a shearing force of 0.5 mm/min until failure.

Preparation for visualization using field-emission scanning electron microscope

Sheared enamel interfaces and the corresponding sheared cylindrical fissure sealant surfaces for a few representative samples were examined using magnifications of up to $\times 4000$ for analysis of surface morphology, with emphasis on areas of adhesive failure or areas of cohesive failure-in-enamel. The specimens were mounted on aluminum stubs with conductive silver liquid, gold sputter-coated and examined under a field-emission scanning electron microscope (SEM) (TESCAN-Vega3, Tescan, Czech Republic) for the verification of the type of failure.

Mode of failure fractured

Modes of failure were examined under an SEM. Failures were categorized as one of the following:

adhesive failure at the enamel and adhesive interface, cohesive failure-in-fissure sealant, cohesive failure-in-enamel, or a mixed failure that includes partial cohesive and adhesive failure.

Statistical analysis

The collected data were analyzed using the SPSS Version 20 (SPSS Inc., IL, USA). Data were analyzed using one-way ANOVA and *post hoc* Tukey's tests for pairwise comparison of SBS values related to the groups of the study.

RESULTS

The SBS data for CPB and RBLC groups are presented in Table 1.

Data analysis with one-way ANOVA revealed statistically significant differences between the groups ($P < 0.001$). Therefore, a *post hoc* Tukey test was used for pairwise comparison.

The results revealed a significant difference between all groups in comparison with the control group ($P < 0.001$) except for Group D. The control group showed significantly lower SBS followed by Group D ($P = 1.0$). The highest SBS was for fluoride-releasing resin-based adhesive (CPB + NaOCl) (Mean SBS = 7.52 MPa). However, the difference between CPB and CPB + NaOCl (with or without pretreatment) was not statistically significant. The second-highest value was related to Group E, in which the specimens were pretreated with NaOCl before applying RBLC. Results showed a marginally statistically significant difference between RBLC and RBLC + NaOCl ($P = 0.049$).

Figure 1 illustrates representative images from the visualization of samples by SEM. The results of SEM correspond with the mentioned data analysis. In Figure 1a (NB group) and D (RBLC) that are related to the groups with the lowest SBS, presence of gap,

Table 1: Mean±standard deviation of shear bond strength of all tested groups

Group	n	Mean±SD of SBS (Mpa)	Minimum (Mpa)	Maximum (Mpa)
A - NB	20	4.34±2.10 ^a	0.95	9.29
B - CPB	20	7.08±2.47 ^b	2.58	12.30
C - CPB + NaOCl	20	7.52±2.74 ^b	2.66	13.10
D - RBLC	20	4.48±1.81 ^a	1.45	7.36
E - RBLC + NaOCl	19	6.24±2.19 ^b	2.58	10.30

Groups identified by different superscript letters were statistically significantly different, and groups with common superscript letters were not statistically significantly different. CPB: Clear fill protect bond; RBLC: Riva bond LC; NB: No bonding; SBS: Shear bond strength; SD: Standard deviation

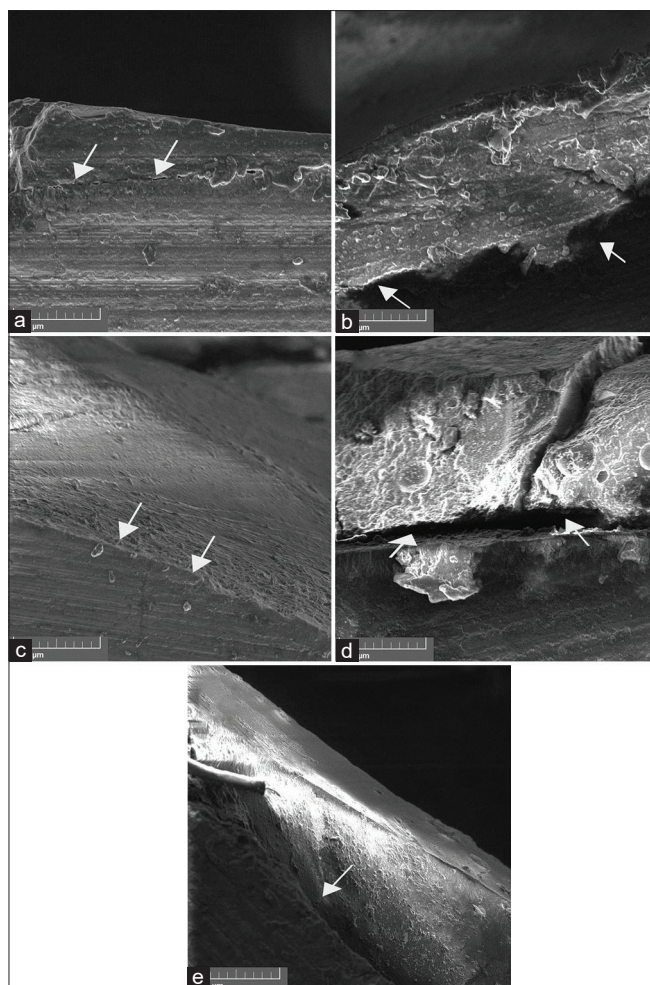


Figure 1: Representative images from the visualization of the specimens by scanning electron microscope. (a) Control group with no bond agent, (b) clear fill protect bond group, (c) clear fill protect bond + sodium hypochlorite, (d) Riva bond LC, (e) Riva bond LC + sodium hypochlorite. Arrows show the gap between enamel and fissure sealant.

and adhesive failure at the enamel and adhesive interface is evident.

DISCUSSION

Despite the proven efficacy of RBPFS, retention is still the main determinant in maintaining the caries preventive effect.^[7] A durable bond between the tooth enamel and fissure sealant is necessary for clinical success of this preventive treatment. Loss of the sealant or partial bond failure can lead to increased risk of carious lesion development due to microleakage and recurrent caries.^[22] Application of an adhesive agent before sealant placement allows optimal infiltration of etched enamel and formation of long adhesive tags. The dehydrating activity of

solvents in the adhesive such as acetone or ethanol displaces water from the deep fissure walls and allows deeper penetration of adhesive and sealant resin. This has been shown to increase bond strength, reduce microleakage, and improve short-term clinical success.^[11]

In agreement with other studies that used bonding agent before RBPFS placement, the present results also showed that applying bonding agent increases the SBS of the RBPFS in comparison to the control group.^[10,11,23,24] Historically, there has been a tendency toward using fluoride-releasing RBPFS materials to decrease the possibility of recurrent caries associated with the sealant, but of special interest is the development of fluoride-releasing adhesive materials. This new type of material shows a more efficient short-term release of fluoride ions as compared with previous resin-based materials.^[12] Therefore, we compared two types of these adhesives (FRSE adhesive and an RMGIC-based adhesive) as an intermediate bonding layer. Based on our results, the FRSE adhesive showed significantly higher SBS than the RMGIC-based adhesive.

The superior performance of self-etch adhesives is due to improving the rheology of the RBPFS, which allows its better flow into the etched enamel. Methacrylate components such as 2-hydroxyethyl methacrylate present in self-etching primers are also responsible for better monomer infiltration.^[1,25]

On the other hand, the composition of enamel (approximately 96% inorganic hydroxyapatite by weight and 4% organic and water component) favors bonding of resin-based adhesives systems over glass ionomer-based adhesives.^[26] In enamel, the degree of organic molecular entanglement between organic tissue fibers and polyacrylic acid molecules cannot be achieved as in dentine. Instead, bonding between enamel and resin-based adhesives is established by the polymerization of monomers inside the microporosities of acid-etched enamel.^[27]

In recent studies, pretreatment of enamel with NaOCl has improved the success rate of fissure sealing. It is probable that NaOCl is causing a reduction in surface stress, allowing the material to penetrate more, increasing its adherence and bond strength on the dental enamel.^[28] Kielbassa *et al.* reported that through liquefaction of organic materials by NaOCl the quality of etching pattern and sealant bond strength will be improved.^[16] Garrocho-Rangel *et al.*

also recommended the deproteinization method before enamel acid etching to obtain better clinical results with sealants.^[29]

The results of our study showed that pretreatment of the enamel with 5.25% NaOCl for 1 min increased the SBS of the RBPFS, but the effect of NaOCl was more prominent and statistically significant when RMGIC-based adhesive was used ($P < 0.001$); similar to results from other researchers.^[27,29] Justus *et al.* reported the lowest SBS for orthodontic brackets when using RMGICs, but after NaOCl pretreatment, RMGICs could be used as an alternative to resin-based adhesives in order to reduce the incidence of white spot lesions.^[30]

The effect of enamel pretreatment with NaOCl on improving the bond strength of RMGIC-based adhesive is also confirmed by SEM images in the present study [Figure 1d and e].

It has been speculated that NaOCl may lead to a mineral surface rich in hydroxyl carbonate and phosphate groups which becomes available for improving chemical bonding of GICs to the enamel.^[31] This mechanism could be considered for RMGIC adhesive as well.

Data about the effect of NaOCl pretreatment on bond strength of self-etch adhesives to dentin tissue are contradictory.^[32,33] However, in contrary to our results researches that used hypochlorite pretreatment before orthodontic bracket attachment to fluorosed enamel reported improved results. The authors attributed the higher SBS values to rougher enamel surface obtained.^[34]

Enamel pretreatment with NaOCl is a cost-effective and innovative technique which deserves further investigation, in the context that it requires careful use in a clinical situation.

CONCLUSION

Using fluoride-releasing resin-based adhesive with NaOCl pretreatment as an intermediate layer gave the highest bond strength of RBPFS to the enamel. Resin-modified GIC-based adhesive with NaOCl pretreatment provided similar bond strengths.

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Conflicts of interest

The authors of this manuscript declare that they have no conflicts of interest, real or perceived, financial or nonfinancial in this article.

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