

Comparative Evaluation of the Effect of Two Different Cleansing Protocols on the Bond Strength of a Self Etching Adhesive to Dentin Contaminated with Hemostatic Agent- An in Vitro Study

Dr. Mallika Shetty¹, Dr. Roshini Anandan^{2*}, Dr. Rajesh Shetty³, Dr. Sanath Kumar Shetty⁴, Dr. Sania Hussain⁵, Dr. Fahad Mohammad⁶

¹*Additional professor, Yenepoya Dental College and Hospital, India,
mallikashetty@yenepoya.edu.in*

²*Post graduate, Yenepoya Dental College and Hospital, India, roshi.anandan@gmail.com*

³*Professor and HOD, Yenepoya Dental College and Hospital, India,
dr_rajeshshetty@yahoo.co.in*

⁴*Professor, Yenepoya Dental College and Hospital, India, drshetty@rediffmail.com*

⁵*Post graduate, Yenepoya Dental College and Hospital, India, sania.aman07@gmail.com*

⁶*Reader, Yenepoya Dental College and Hospital, India, fahadma@yenepoya.edu.in*

OBJECTIVES: Gingival retraction using impregnated retraction cords using hemostatic agents like 25% Aluminium Chloride is the most important step in isolation of bonded restorations. These agents are known to hinder the bonding of resin to dentin. The aim of the study was to evaluate the effect of two different cleansing protocols on the bond strength of a self etching adhesive to dentin contaminated with hemostatic agents. **METHODS AND MATERIALS:** Sixty three 1st premolars extracted for therapeutic reasons were randomly divided into three groups A, B and C of 21 samples each. The mid coronal dentin was exposed in all the samples and an even smear layer was produced using a standard diamond point and sandpaper grit. All the samples were contaminated with 25% Aluminium Chloride hemostatic gel. Group A was decontaminated with only water spray, Group B with water spray and 2 % Chlorhexidine (CHX) and Group C with water spray, 2% Chlorhexidine and 10% EDTA gel. All the samples were bonded using a self etch adhesive and composite resin. The microshear bond strength was assessed using Universal Testing Machine (UTM). **RESULTS AND DISCUSSION:** A significant improvement in the microshear bond strength was noted in Group B(Water spray and 2%Chlorhexidine) when compared to the Control group A and Group C. **CONCLUSION:** Decontamination of dentin with water spray and 2% Chlorhexidine solution significantly improved the bond strength of self etch adhesive restorations.

Keywords: Aluminium Chloride, Chlorhexidine, EDTA, Gingival retraction, Self etch adhesive.

1. Introduction

Gingival retraction using chemically impregnated retraction cords and hemostatic agents for isolation is a common practice in bonded restorations. Commonly used commercially available hemostatic agents used for gingival retraction are Aluminium chloride and Ferric sulphate.¹ Bonding to dentin is usually the combination of the characteristic smear layer formed during tooth preparation and the effect of the etchants on the smear layer. Adhesion to dentin is achieved by using either etch and rinse or self etch approach.² Self etch adhesives have started gaining more importance because of the relative ease of use and less destruction of the collagen fibers of the dentin. The etchants used in the self etch adhesive groups are relatively weaker acids compared to the total etch group.³

Gingival retraction agents are in contact with the tissues and the dentin usually between 4-10 mins.⁴ Hemostatic agents predominantly Aluminium chloride and Ferric sulphate alter the dentin smear layer and makes it resistant to etching caused by the bonding system.^{4,5} Hence it is vital to formulate a cleansing protocol to remove any residual hemostatic agent present in the dentin prior to bonding. ^{5,6,7}

There is no standard protocol recommended for decontamination of dentin after use of hemostatic agents or comparison of these cleansing protocols and their effect on the bond strength of self – etch adhesives. Thus the study aims to evaluate the effect of two different cleansing protocols on the bond strength of a self etching adhesive to dentin contaminated with hemostatic agent.

2. Methods

The in-vitro experimental study was conducted in Mangalore, Karnataka, India for a period of 12 months. Ethical clearance was obtained from the University ethical committee prior to sample collection. The sample size was calculated using the G + software. Strong effect of magnitude 0.8 was assumed between CHX and water spray and CHX and EDTA. In order to detect the anticipated effect with 5% level of significance, 80% power, 21 samples were included in each group. The total sample size was 63. The sampling method followed was simple random sampling.

PROTOCOLS FOLLOWED

Preparation of the dentin samples and contamination with 25% Aluminium Chloride:

Sixty three 1st premolars extracted within 3 months of the study period for periodontal and orthodontic reasons were included for the study. Premolars with caries or any other dentinal anomalies and gross dentinal fractures were excluded from the study. After extraction the collected teeth were cleaned thoroughly using brushes and currettes and stored in 1% chloramine solution (Figure 1).

The dentin samples were embedded in cold cure acrylic blocks (DPI® HEAT CURE) – (ISO 20795-1 : 2013) measuring 3x1.5 cm. The mid coronal dentin of the samples was exposed by using a low speed diamond grit with water coolant following which the dentin surface were polished using a 600 grit sandpaper to obtain an even smear layer for bonding. ^{6,9} (Figure 2). All the specimens were treated with 25% Aluminium Chloride (Viscostat clear™) for 4

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mins(Figure 3).

The specimens were grouped randomly into 3 groups – Group A, Group B and Group C of 21 specimens each respectively . In Group A (Control group - only water spray was used for 60 seconds and air dried with short bursts of air for 30 seconds . In Group B (Decontamination with water spray + 2% Chlorehexidine solution) the specimens were decontaminated following water spray with 5ml of 2% CHX for 1 minute and rinsed with water for 60 seconds. In Group C (water spray + 2% CHX + 10 % EDTA) the specimens were decontaminated following water spray with 5ml of 2% CHX for 1 minute and rinsed with water for 60 seconds and 10 % EDTA for 60 seconds and rinsed with water for 60 seconds

All the samples were treated with single bond self adhesive agent for 35 seconds followed by air spray for 10 seconds. Light cure composite resin (Z 250 Filtek) (3M ESPE, St. Paul, MN, USA) was applied into prefabricated tubes of increments 1 mm twice and was light cured using a LED light curing unit .

Measurement of microshear bond strength:

All the specimes were tested for debonding using Universal testing machine. The microshear bond strength was measured at a crosshead speed of 0.5mm/min.⁶ The bond strength was measured in megapascals (MPa)

Statistical analysis was done using the SPSS software. One-way ANOVA was used to compare the microshear bond strength of the 3 groups A, B and C. Post Hoc test was used for multiple comparison

3. Results

A total of 63 freshly extracted premolars were tested in the current study. The results revealed the highest mean microshear bond strength of 557.9 ± 41.2 Mpa for Group B (Decontamination with 2% CHX) followed by Group C (Decontamination with 2% CHX followed by 10% EDTA) with a mean of 392.7 ± 36.0 Mpa . The least microshear bond strength observed was in Group A (control group- no decontamination) with a mean shear bond strength of 286.6 ± 29.9 Mpa ($P < 0.001$) (Table 1) .

Further intergroup comparison was done between the three tested groups. Group B had the highest micro shear bond strength when compared to both Group A and Group C individually. The mean difference was statistically highly significant ($p < 0.001$). Intergroup comparison between Group C and Group A also was statistically highly significant with the highest mean value for Group C. ($p < 0.001$) (Table 2) .

4. Discussion

The study evaluated the effect of two different cleansing protocols using CHX and EDTA on the shear bond strength of composite resin. Bonded restorations have gained great importance in fixed prosthodontics with the improvements in the bonding protocols.¹⁰ However isolation and presence of a clean uncontaminated dentin surface is critical to achieve success of bonded restorations. Marginal seal in bonded restorations are of a great importance and can be

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hampered if the marginal dentin bond is compromised .

Various studies have concluded that the use of 25 % Aluminium Chloride gel for gingival retraction alters the dentin smear layer and thus impairs the bonding of dentin to resin based cements or restorative materials.^{1,3,5,7,9} The use of universal adhesive agents for bonding following use of these retraction agents have increased risk of compromised bonding , also various dentin decontamination agents have been studied in the recent years to enhance the bonding.³ Literature evidence suggests that Aluminium chloride affects the dentin bonding by 2 major mechanisms

- Highly acidic ph (0.5-0.7) disturbs the dentin smear layer and hybrid layer
- Replacement of Ca by Al in the dentin hydroxyapatite crystals which makes the dentin resistant to etching.⁶

The study investigated the effect of decontamination of dentin using 2% Chlorhexidine and 10% EDTA. The microshear bond strength was the highest for Group B (557.9 ± 41.2) , where decontamination was done using water spray and 2% Chlorhexidine. Chlorhexidine is a cationic biguanide detergent with strong antimicrobial properties. Various studies have demonstrated the use of CHX as a cavity and root canal disinfectant.¹¹

The findings of present the study were in agreement with the findings of the study conducted by Saati et al in the year 2020 , but the study was conducted using an etch and rinse technique for bonding.⁶ The study revealed that, the use of Chlorhexidine significantly improved the microshear bond strength of dentin to composite resin. The mechanism of action of CHX as a decontamination agent has been widely studied.^{5,6,8}.

The improvement in the microshear bond strength of dentin followed by application Chlorhexidine is probably due to two actions. CHX is a potent antimicrobial agent and detergent, hence it decontaminates any residual free unbound aluminium chloride particles in the dentin surface.^{12,13} Secondly, CHX is a potent MMP (Matrix Metalloproteinase) inhibitor. Endogenous MMPs causes degradation of the collagen fibrils in the dentin hybrid layer, causing bond degradation. CHX effectively inhibits the MMPs by restoring the disturbed dentin hybrid layer by the high ph of the hemostatic agent.^{13,14}.

10% EDTA was also used to decontaminate the dentin. EDTA because of its strong chelating effect has also been widely studied to restore the bond strength of dentin . However in the present study, it was used in combination with 2 % Chlorhexidine (Group C) . The intergroup comparison revealed that the microshear bond strength after using only Chlorhexidine was significantly higher than the microshear bond strength after using 2% Chlorhexidine and 10% EDTA . The finding of the study was contradicting the results of a previous studies conducted by Ajami et al in 2013¹⁵ and Singh et al in 2015.¹⁶

The negative effect of EDTA on the bonding of dentin might be due to 1) EDTA was used in combination with CHX, which probably retarded the effect of CHX, 2) The concentration of EDTA used was 10 % which is less than 0.5 M of EDTA. A study conducted by Sebold et al in 2017 ¹⁷ investigated the effect of EDTA conditioning on bond strength of dentin, where they concluded that high concentration of EDTA of more than 25% is required for the chelating action of EDTA on the dentin smear layer.

However the use of EDTA and CHX was still superior to the control group when an intergroup comparison was done between Group A and Group C. This is probably due to the strong decontamination action of CHX. CHX remains to be the confounding factor in both the test groups.

The findings of the study have some limitations, as the study was done in-vitro, hence the exact dentin bonding cannot be mimicked. Also the samples were not thermocycled prior to bonding, hence the long term effect of dentin bonding cannot be conclusively studied

5. Conclusions

Within the scope of the current study, it can be concluded that

1. Use of 25% Aluminium Chloride significantly reduced the dentin bond strength
2. Use of only water spray is insufficient to decontaminate the dentin
3. Decontamination with water spray with 60 secs followed by use of 2 % CHX solution for 1 minute significantly improves the bond strength of dentin to resin
4. Use of 10% EDTA as a dentin conditioning agent along with 2 % CHX has a negative effect on the shear bond strength

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