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THE EFFECT OF SODIUM HYPOCHLORITE TREATMENT ON THE SHEAR BOND STRENGTH OF AN ACETONE-BASED ADHESIVE SYSTEM TO DENTIN – AN IN VITRO STUDY

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ABSTRACT

Aim: The aim of the present study was to evaluate the effects of different concentrations of sodium hypochlorite applied for 30 seconds on acid etched dentin on the shear bond strengths of an acetone-based adhesive. **Materials and Methods**: 40 freshly extracted molars were used as specimens to evaluate shear bond strength of composite to sodium hypochlorite (NaOCl)-treated dentin using Prime and Bond NT dentin bonding agent after 10% NaOCl (Group I), 5% NaOCl (Group II), 2.5% NaOCl (Group III) and No NaOCl (Control) treatment. Shear bond strengths of all specimens were measured using an Intron Universal Testing Machine. **Results**: Group II demonstrated higher bond strengths than Groups I, III and IV. Groups I, III and IV did not show any statistically significant differences in their bond strengths. Group III demonstrated the least bond strength. Group I showed similar bond strength as that of Group IV. **Conclusion**: Highest shear bond strength values were demonstrated by Group II i.e. 5% sodium hypochlorite treatment group. This could be because of partial decollagenation and formation of an optimum hybrid layer.

KEY WORDS: Acetone, Dentin adhesive, Dentin Bonding, Hybrid layer, Shear bond Strength, Sodium Hypochlorite

INTRODUCTION

An ideal restorative material is one, which adheres to enamel and dentin and mimics all the properties of the dental tissues as closely as possible. This ensures an esthetic and highly functional restoration.[1] Clinical success of composite resins depends on effective bonding to enamel and dentin.[2] Adhesion of resin to enamel is highly predictable and durable due to the micromechanical bond between the resin bonding agent and the highly inorganic substrate of enamel. However, bonding of composite resins to dentin is comparatively difficult due to the following reasons- (1) complex structure of dentin with a low inorganic content randomly arranged in an organic collagen matrix (2) presence of dentinal fluid, which is constantly flowing outwards and (3) proximity of dentin t o pulp.[3,4]

Early dentin bonding agents attempted at bonding either the inorganic or organic constituents of dentin.[5] The smear layer was preserved, thus reducing dentin permeability and fluid seepage out of the dentinal tubules. The dry surface created provided increased micro porosities for bonding hydrophobic resin materials. However, the bond strengths achieved were poor and the bond was not durable.[6] Since the early 1990s, there has been rapid progress in the development of new dentin adhesive systems. Originally adhesion to dentin involved several steps with separate components for priming and bonding. Later one-bottle dentin adhesive systems were developed. The current basis for dentin bonding is the formation of a hybrid layer resulting from resin penetration into the acid demineralized dentin. Acid etching removes the smear layer and demineralizes the superficial dentin, thus exposing the collagen fibrils.[7]

Application of the dentin-bonding agent causes enmeshment of the exposed collagen fibrils by hydrophilic resin monomers. This is the resin-dentin hybrid layer located between the dentin and the composite restorative material.[8] Several in vitro studies have indicated that the

Fig.1 All Groups (40 Samples)

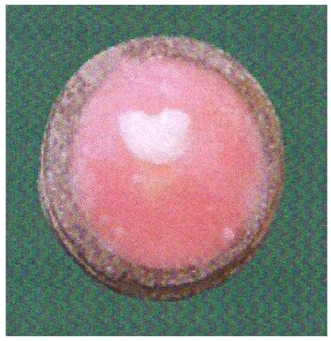

Fig.2a Tooth Mounted with Buccal/Lingual Surface Facing

Fig.2b Surface Flattened with Carborundum Disc to exposed dentin

Fig.2cSample Checked for Parallelism on Surveyor

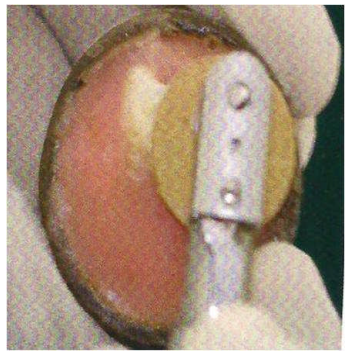

Fig.2d Tooth Polishing with sand Paper

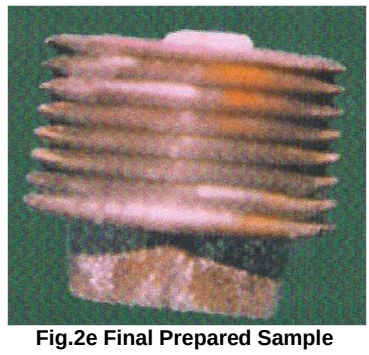

Fig.2e Final Prepared Sample

hybridization of the dentinal tubules and inter tubular dentin significantly enhances the bonding of the resin to dentin.[9,10,11,12] However, exposure of the collagen network on etched dentin represents a soft, delicate bonding substrate that makes dentin bonding procedures highly technique sensitive.[13,14]

To make bonding to dentin as predictable as enamel bonding, several researchers have suggested complete removal of the collagen network on the etched dentin surface before bonding.[15,16] Various agents have been recommended for de-proteinization of the etched dentin. Of there, sodium hypochlorite has been reported to be the most effective agent by many researchers.[17,18] Studies have reported the use of concentrations varying from 1.5% to 10% sodium hypochlorite, for time intervals of 30, 60 and 120 seconds.[19,20,21,22]

The aim of the present study was to evaluate the effects of different concentrations of sodium hypochlorite applied for 30 seconds on acid etched dentin on the shear bond strengths of an acetone-based adhesive.

Materials and Methods

Sample Selection:

Forty freshly extracted molars were used as specimens to evaluate shear bond strength of composite to sodium hypochlorite (NaOCl)-treated dentin using Prime and Bond NT dentin bonding agent after 10% NaOCl, 5% NaOCl, 2.5% NaOCl and No NaOCl treatment. The teeth were divided into 4 groups, each containing 10 samples **(Fig.1)**.

1. Group I: 10% NaOCl + Primeand Bond NT
2. Group II: 5% NaOCl + Primeand Bond NT
3. Group III: 2.5% NaOCl + Primeand Bond NT
4. Group IV (Control Group): Primeand Bond NT alone

Preparation of Samples:

Each tooth was embedded into cold cure acrylic (DPI-RR cold cure-IS 6887, Type II Class I) contained in a threaded metallic coping with 3.5 inch external diameter, 2.5 inch internal diameter and 1.0 inch height with the buccal or lingual surface of the extracted teeth facing up

and kept slightly above and parallel to the outer surface of the coping **(Fig.2a)**. A flat area was created on the exposed dentinal surface about 4mm in diameter with a carborundum disc **(Fig.2b)**, maintaining it at 90° to the tooth surface so that a surface parallel to the coupling is created. The parallelism of the surface was evaluated on the surveyor and then adjusted and rechecked on the surveyor **(Fig.2c)**. The dentinal surface was smoothened with fine grit sand paper discs **(Fig.2d)** till a shiny dentinal surface was achieved **(Fig.2e)** and the sample was stored in distilled water at room temperature.

The prepared dentinal surface was acid etched with 3M etchant for 15 seconds (**Fig.3a**), rinsed with water for 15 seconds and blot dried with blotting paper (**Fig.3b**). 10% NaOCl (Group I), or 5% NaOCl (Group II), or 2.5% NaOCl (Group III), or No NaOCl (Group IV) was applied on the dentinal surfaces for 30 seconds (**Fig.3c**) and rinsed with water for 15 seconds and blot dried (**Fig.3d**) leaving visibly a moist dentin. Prime and Bond NT adhesive was applied and cured for 20 seconds **(Fig.3e)**. The plastic tubing was placed on the bonded dentin surface; Z-250 composite was packed into it in two increments and light cured for 20 seconds each **(Fig.3f)**. Following this, the tubing was removed and shear bond strength was evaluated in an Intron Universal Testing Machine (**Fig.4**).

Preparation of 10% Sodium Hypochlorite Solution:

10% sodium hypochlorite solution (**Fig.5**) was prepared in the Biochemistry laboratory and it was diluted to 5% and 2.5% by taking 1:1 ratio by volume of sodium hypochlorite and distilled water.

Statistical Analysis:

The statistical analysis was done using Kruskal Wallis test and multiple comparisons using SPSS software.

Results

Shear bond strengths of all the samples tested and their means along with standard deviations are summarized in **Table I**. Results of statistical analysis are given in **Table II**. The following can be derived.

1. Group II demonstrated higher bond strengths than Groups I, III and IV.
2. Groups I, III and IV did not show any statistically significant differences in their bond strengths.
3. Group III demonstrated the least bond strength.
4. Group I showed similar bond strength as that of Group IV (control group).

Discussion

Achieving consistent adhesion of composite resin to tooth structure possess a major challenge in restorative dentistry. A durable bond between composite resin and tooth structure has several clinical advantages such as:

Fig.3a Acid Etching for 15 Seconds

Fig.3b Washed and Blot Dried

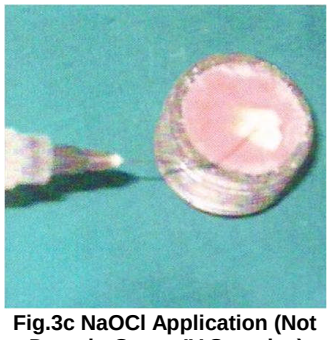

Fig.3c NaOCl Application (Not Done in Group IV Samples)

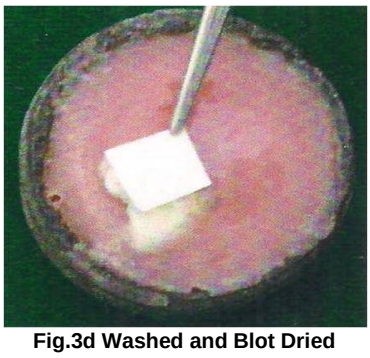

Fig.3d Washed and Blot Dried

Fig.3e. Bonding Agent Applied and Cured

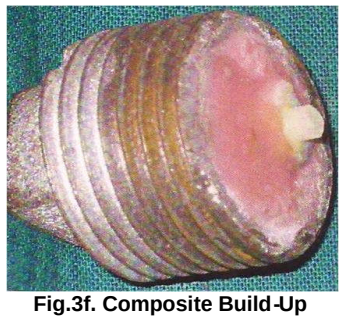

Fig.3f. Composite Build-Up

Fig. 4 . Intron Universal Testing Machine

Fig.5. 10% Sodium Hypochlorite Solution

(1) reduced micro leakage (2) improved strength of restored teeth and (3) reduced stresses due to polymerization shrinkage of resin.[23]

Predictable adhesion to enamel has been possible since 1955 when Buonocore introduced the acid etch technique.[24] However, adhesion of composite resins to dentin has been an elusive goal until recent times. This is because of the inherent nature of dentin that has a high organic content, variable structure and the constant flow of dentinal fluid.[15,25] To solve this problem, an intermediate material – dentin bonding agent – was developed having both hydrophilic and hydrophobic properties, which enables it to bond to the dentin and composite resin respectively.[21]

The early dentin bonding agents retained the smear layer and attempted at chemical bonding to dentin. However, their bond strengths were poor and hence they were not clinically successful.[15] Since the 1990s, several advancements have been made in dentin bonding agents. One concept is the "total-etch concept" of enamel and dentin simultaneously. While this produces micro porosities on the enamel surface, it simultaneously removes the smear layer on the dentin and exposes the collagen fibrils.[21] Modern dentin bonding agents contain hydrophilic monomers as primers along with a solvent such as acetone or ethanol and an adhesive resin.

Following acid-etching of dentin, the solvent dries away the dentinal fluid, while the hydrophilic monomers enhance the infiltration of the bonding resin into the exposed collagen network and the dentinal tubules, to form the "hybrid layer" or the "resin-dentin inter-diffusion zone".[9] However, acid-etch technique is highly technique-sensitive. After demineralization of dentin, water occupies the inter-fibrillar spaces left by solubilization of the mineral phases and maintains the collagen network in an expanded state. If dentin is over dried, the water evaporates from the inter-fibrillar spaces causing collapse of the collagen network. Conversely, excess moisture on the dentin surface can result in poor penetration of the dentin bonding agent and therefore poor bonding to dentin.[23] This will impede the penetration of the adhesive monomers and formation of the hybrid layer with a consequent drop in bond strength

To overcome these problems, Gwinnett and Tay recommended the "moist-bonding concept". Here, the dentin surface is left visibly moist after etching and rinsing prior to application of the dentin-bonding agent. This would prevent the collapse of the collagen fibril network and therefore enhance the bond strength to dentin.[27] However, concerns have been raised that most modern dentin bonding agents do not fully diffuse through the collagen network that remains after acid-etching of dentin. This leaves a zone of unsupported collagen fibrils beneath the

Table.1 Bond Strengths of All Sample Teeth in Mpa

Sample	*Group I*	*Group II*	*Group III*	*Group IV*
1.	21.42	27.95	17.96	32.78
2.	25.45	27.48	22.31	14.42
3.	15.14	45.12	20.29	17.15
4.	14.17	18.36	17.07	16.43
5.	23.44	34.63	17.40	20.38
6.	26.66	23.36	16.11	22.07
7.	13.21	27.56	25.61	12.00
8.	16.99	20.21	17.88	24.72
9.	37.53	24.00	17.83	22.31
10.	12.89	18.85	14.43	14.58
Mean ± SD	20.69±7.8	26.75±8.1	18.68±3.2	19.68±6.1

Table.2 Kruskal-Wallis Test

	n	*Mean Rank*	*Chi Square*	*Df*	*'p' Value*
Group I	10	18.2	8.157	3	0.043*
Group II	10	29.6			
Group III	10	16.75			
Group IV	10	17.45			

** Statistically significant*

hybrid layer, which is not infiltrated by the dentin-bonding agent.[12,13]

. Sano *et al*. have reported that these bands of exposed collagen, not protected by the adhesive resin, would undergo hydrolysis with long term exposure to oral fluids. This results in "nano leakage" and eventually the bond between the composite resin and dentin fails.[16] In the light of this evidence, several researchers have suggested that the presence of collagen fibrils is questionable for the bond strength of composite resins to dentin.[12,13,15] Gwinnett reported that removal of collagen layer might be beneficial for bonding the composite resin to dentin.[8] This has been supported by other researchers.[23,28,29] Many agents have been recommended for the same, and of these, sodium hypochlorite (NaOCl) shows the most promise.

NaOCl is a non-specific proteolytic agent that effectively removes organic compounds at room temperature. Tanaka *et al*. demonstrated that the use of NaOCl for surface treatment of etched dentin enhanced the shear bond strengths of dentin bonding agents to dentin.[18] Saboia *et al*. reported that NaOCl enhances the bond strengths of acetone-based adhesives while ethanol-based adhesives exhibited a significant reduction in bond strengths.[29] NaOCl deproteinization increases the surface roughness of dentin and its wettability. It exposes a labyrinth of lateral, secondary tubules, which allows good mechanical retention. This substrate is also rich in exposed hydroxyapatite crystals and may result in a stable interface over time.[30] Inai *et al*. have reported that acetone containing adhesives interact strongly with a treated dentinal surface that has a high mineral content.[31]

In the present study, an acetone-based adhesive – Prime and Bond NT – was employed. This is a nanofilled adhesive containing cetylamine hydrofluoride as the filler. Research has also showed that nanofillers also infiltrate deep into the dentin along with the resin monomer and thus contributes to the bond strength.[20] The composite resin employed was Filtek Z-250 (3M ESPE), a micro hybrid, universal restorative material. Shear bond strength was checked in this study to simulate the oral conditions for restorative material to fracture on loading.

According to the results of this study, there was no statistical difference between 10% NaOCl, 2.5% NaOCl and the control groups. This could be explained by the observation that 10% NaOCl applied for 30 seconds after acid etching could have resulted in complete depletion of collagen, which causes minimal or no hybrid layer formation. This is in accordance with the findings of Chersoni S *et al*.[32]

Some researchers have reported that the formation of a hybrid layer is not so important for high shear bond strengths of dentin bonding agents.[15,33] However, Ferrari *et al*.[34] and Perdigao *et al*.[22] have reported that complete depletion of collagen leads to minimal or no hybrid layer formation, which could decrease the retention of the restoration. The findings of the present study support the findings of their investigations.

In the present study, Group III samples exhibited less shear bond strengths similar to that of the control group. Although 2.5% NaOCl is equivalent to 5% NaOCl in its collagen dissolving ability, it may be inferred that the application time may not have been adequate to produce hi8gh shear bond strengths. The shorter application time may have caused insufficient deproteinization of dentin. Hence, it may be hypothesized that a longer application time would be beneficial while using a 2.5% solution of NaOCl. 5% NaOCl applied for 30 seconds after acid etching shows the highest shear bond strengths in this study. Earlier studies by Vargas MA *et al*.[35] and Perdigao *et al*.[36] using 5% NaOCl for deproteinization reported the absence of hybrid layer following bonding when the application time ranged from 1 to 2 minutes. However, in the present study, the application time was only 30 seconds. It may be therefore hypothesized that, by reducing the application time of 5% NaOCl, complete deproteinization of the etched dentin does not occur. There may be partial decollagenation of the dentin with remnants of collagen fibrils in the deeper portion of the etched dentin. Perdigao *et al*.[22] have reported that the integrity of collagen fibrils left unexposed upon acid-etching still has a significant role in achieving good bond strengths. Probably 5% NaOCl treatment for 30 seconds may have retained some remnants of collagen fibrils which could enhance the intermingling of adhesive monomers with the filigree of collagen fibrils, thus forming an optimum hybrid layer. For the control group, decreased shear bond strengths were obtained. Probable reason for this could be due to the presence of a thick unsupported collagen layer not properly infiltrated by the adhesive resin.

The bonding agent employed in this study was Prime and Bond NT. Prati C *et al*.[14] reported that the acidic monomers in this agent may re-etch the mineral phase of the partially deproteinized dentin to form a nano-hybrid layer. The presence of nanofillers in the bonding agent may also be a factor in establishing high bond strengths.

Though the results of the present study cannot be directly related to the clinical situation, it provides valuable information regarding dentin pre-treatment using NaOCl for acetone-based adhesives. Independent, long-term clinical studies are necessary to validate the findings of this study.

CONCLUSION

The following conclusions can be drawn from this in vitro study-

1. Sodium hypochlorite pre-treatment is recommended for enhancing shear bond strength of acetone-based adhesives.
2. 10% sodium hypochlorite treatment exhibits lower shear bond strength than 5% sodium hypochlorite treatment. However, it demonstrates slightly better bond strengths than 2.5% sodium hypochlorite and no hypochlorite treatment.
3. Highest shear bond strength values are demonstrated by 5% sodium hypochlorite treatment group. This could be because of partial decollagenation and formation of an optimum hybrid layer.
4. 2.5% sodium hypochlorite treatment demonstrates the least shear bond strength values; probably longer application time was necessary to produce high shear bond strengths.
5. Formation of hybrid layer may be important for achieving good shear bond strengths.
6. Long-term clinical studies are necessary to validate the findings of the present study and its significance in clinical practice.

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