Evaluation Method of Bonding To Root Canal Dentin After Sodium Hypochlorite Treatment*

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ABSTRACT

Statement of the problem: Shear bond strength test is a common test to evaluate the bond between restorative materials and tooth structure but it cannot measure the marginal adaptation of the restorative filling materials. Therefore, in this research an evaluation method of punch out test was employed to evaluate resin bonding to root canal wall dentin by measuring both marginal adaptation and shear bond strength simultaneously.

Objectives: The aim of this study is to investigate the reliability and efficiency of punch out shear test to evaluate the effect of sodium hypochlorite (NaOCl) as a root canal irrigant on composite resin bonding to canal dentin and also to evaluate the contraction gap formation from the same specimen. The null hypothesis tested was that treatment of root canal with NaOCl would not compromise bonding to root canal dentin, whether a total etching adhesive or self-etching adhesive was used.

Materials and Methods: A total etching adhesive system (Optibond Solo Plus) and a self etching adhesive system (XenoIII) were employed in this study. A standardized truncated cone cavity (4 mm in base diameter, 3 mm in depth and a convergence angle of $5°$), was prepared at the top portion of the root canal. The cavities were subjected to sodium hypochlorite treatment for different periods of time and then rinsed with distilled water. Then, one of the aforementioned dentin bonding systems was applied to the cavity walls according to the manufacturer's instructions. The cavities were then filled with a micro-hybrid composite resin.

Results and conclusion: The increase in the NaOCl application time resulted in a progressive decrease in the punch out shear bond strength for the specimens treated with a self etching adhesive system. A marginal gap was detected when XenoIII was applied after NaOCl application.

Key words: Root canal dentin, self adhesive, sodium hypocholorite, punch out sher bond strength.

INTRODUCTION

The most appropriate kind of mechanical testing regime for evaluating restorative materials has not been agreed amongst the international community responsible for developing standard tests for these products. Recently, a shear punch test has been advocated[1] as it has certain advantages over more traditional compressive and flexural tests. The shear punch test has previously been used in standards testing.[2-3]

Restorations for endodontically treated teeth are designed to replace the missing tooth structure and to protect the remaining structure from fracture. Guzy et al.[4] reported that preservation of the structure of endodontically treated teeth is an important factor to prolong the longevity of these teeth.[4] In addition, it is essential to minimize the amount of tooth structure

loss for a favorable prognosis.[5] A good performance of the resin bonding system to the tooth structure is essential to restore the tooth after root canal treatment.[6]

The reliability and predictability of resin bonding to enamel are well established. However, those to dentin are still questionable because dentin has greater organic contents than enamel.[7] Moreover, both the composition and morphology of deep dentin are different from those of superficial dentin. For example, the hardness of deep dentin is less than that of superficial dentin;[8] the density and diameter of dentinal tubules of deep dentin are greater and thicker, respectively, compared with superficial dentin. The calcification of deep dentin is not as mature as that of superficial dentin.[6] Therefore, bond strength to deep dentin has been reported to be lower than that of superficial dentin.[9-10] In addition, deep dentin and root canal wall dentin might be affected by root canal irrigants and disinfectants during endodontic treatment. Sodium hypochlorite (NaOCl) is one of the most common root canal irrigants used for debridement lubrication, destruction of microbes and dissolution of organic tissues.[11-12]

Several researchers[13-14] have studied the role of NaOCl in dentin permeability and dentin adhesion. Depending on each testing methodology and/or specific composition of each dentin adhesive, the application of NaOCl as a

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root canal irrigant may increase or decrease bond strengths of composite resin bonding to tooth structure.[15-20] Nikaido and Perdigao[16-17] reported that NaOCl treatment interfered with the adhesive of the bonding system to dentin and compromise composite bonding. Whereas Inoue et al.[21] reported that NaOCl treatment improved the adhesion of the bonding system to dentin when phosphoric acid etchant is used to remove the smear layer.

The previous studies investigating the effect of NaOCl on resin bonding systems to tooth structure have produced controversial results. Therefore, the effects of NaOCl treatment on dentin have not clearly been confirmed. The shear punch test has previously been used in standards testing[2] and has been suggested as a suitable method for evaluating dental cements.[22-23]

In this study, an evaluation method for resin bonding to root canal dentin was conducted which measures both marginal adaptation and punch out shear bond strength simultaneously using the same specimen. The reliability and efficiency of this method were investigated in an effort to examine the effects of NaOCl as a root canal irrigant on resin bonding to root dentin when a total etching adhesive or a self-etching adhesive was used.

MATERIALS AND METHODS

One hundred extracted bovine mandibular incisors were used in this study. The teeth were stored in 0.5% chloramines solution at 4°C. Roots with a standardized dimension were only selected. Two dentin bonding systems; a self-priming adhesive system containing both the primer and adhesive in one bottle, utilizing the total etching technique (Optibond Solo Plus) and a self-etching adhesive system (Xeno® III), were employed in this study as shown in Table 1.

Cavity preparation

The teeth were cleaned of debris with periodontal curettes and stored in distilled water. The crowns of the teeth were removed with a diamond disk at the cement-enamel junction. The pulp tissues were removed using a dental file (#30, Maillefer, Ballaigues, Switzerland) without root canal preparation. Each root was embedded in the center of an acrylic pipe (diameter: 16 mm, depth: 25 mm) with a chemically cured acrylic resin, Trayresin (Dentsply Trubyte, York, PA). A 3 mm thick coronal surface was resected with a low-speed diamond saw (Isomet 2000, Buehler, Lake Bluff, IL, USA). Then, the root canal space was filled with a thermoplasticized gutta-percha delivery system (Obtura H, Obtura, Fenton, Mo, USA) and a resin sealer (AH-26; DeTrey, Zurich, Switzerland). A standardized

truncated cone cavity[23] (4 mm in base diameter, 3 mm in depth and a convergence angle of 5°), was prepared at the top portion of the root canal using a milling apparatus(KAVO EWL,K9-Milling apparatus Type 990) and a tapered end drill 4mm in diameter (040, Hager&Meisinger GmbH) under a water coolant (Fig.1). The long axis of the drill was directed parallel to the long axis of the root canal. All cavities were rinsed with distilled water using a three-way syringe.

Preparation of surface for adhesion

5% solution of sodium hypochlorite was selected as a root canal irrigant. Its effects on the aforementioned dentin bonding systems to root canal dentin were examined. Specimens were randomly assigned to 2 major groups of 50 teeth each according to the adhesive systems employed and then each main group was subdivided to five subgroups according to NaOCl treatment as follows:

> Group A, neither NaOCl treatment nor bonding system;
> Group B, bonding system without NaOCl treatment;
> Group C, NaOCl for 1 min and bonding system;
> Group D, NaOCl for 5 min and bonding system;
> Group E, NaOCl for 10 min and bonding system.

After a designated period of NaOCl treatment, each cavity was rinsed with distilled water for 10s and one of the aforementioned dentin adhesive systems was applied according to a certain application protocol as mentioned in table 2.

After application of different adhesive systems to cavity walls, a hybrid composite resin (Z250, 3M) was filled in the cavity and photo-polymerized using a light-curing unit (Elipar 2500, 3M ESPE, St Paul, MN-Germany) for 40 s.

Observation of marginal adaptation and gap

Immediately after the resin filling, the tooth surface in the acrylic pipe was ground flat with a series of silicon carbide abrasive paper (#320, #500 and # 1000) under running water. After polishing, the specimens of each test group were immersed for 24 hours at 37°C in a 0.2% fuchsine solution. Then, specimens surfaces were rinsed with distilled water and dried prior to the microscopic examinations.

Marginal gap of the composite inside root canal dentin was investigated with a Nikon, stereoscopic Zoom Microscope SMZ/1000 equipped with digital camera DXM1200F at 20 x magnification. A software version 2.63 (Nikon Corporation, Japan) was installed for measurement of the gap. When fuchsine penetration was observed anywhere around the margin of a specimen, it was defined as a gap positive.

Measurement of punch out shear bond strength

After the contraction gap observation, all specimens were kept in distilled water at 37°C for 24h. A superficial 2.0 mm coronal layer of each specimen was sectioned with a low-speed diamond saw (Isomet 2000, Buehler, Lake Bluff, IL, USA). The thickness of the sectioned specimen and the diameter of the resin-filled cavity were measured using a digital micrometer (Fowler, Cole-Parmer Instrument Co.) with 0.001 mm precision.

Each sliced specimen was positioned and centered on a metal platform for the punch out shear bond strength test (Fig. 2, 3). The load was applied by a cylindrical steel plunger of 2.8 mm diameter attached to the upper jig of the universal testing machine. The metal platform had a round central hole (5.0 mm in diameter) which is larger than the root canal diameter. The most coronal side of the specimen is placed downwards. The load was applied by the plunger from the apical side of the specimen. The plunger was centered over the composite filling inducing shear stresses along root dentin/composite interface. The initial dislodgement load by which the filled resin was punched out from the root canal wall cavity was measured using a universal testing machine (Instron testing machine 8500; Norwood, MA, USA) at a crosshead speed of 1.0 mm/min. In order to express the bond strength (ó) in MPa, the load at failure recorded in Newton was divided by the area of the bonded interface, which was calculated through the following formula: ó = F/A

Where, F = load for specimen failure (N).
A = bonded area (approximately 27 mm^2).

To calculate the bonded area (A), a formula was applied to calculate the lateral area of the conical geometric figure of prepared cavity in the root canal that formed a circular straight cone trunk of parallel bases. The formula used for the area calculation was:
A = πXgX (R1 + R2)

Where, π= 3.14, g = trunk generatrix, R1= smaller base radius, R2= larger base radius (Fig.4).

For the conical trunk generatrix (g) calculation the Pythagorean theorem (the square on the Hypotenuse is equal to the sum of the squares on the other two sides) was used as expressed in the following formula: $g^2 = h^2 + [R2 - R1]^2$

Where, g = conical trunk generatrix, R1 and R2 were obtained by measuring the internal diameters of the smaller and larger base, respectively, corresponding to the internal diameter between the root canal walls.

Statistical Analysis

Occurrence of the gap formation was analyzed using the chi square test at a significant level of 5%. The mean punch-out shear bond strength values from each specimen were initially calculated. Considering each group was composed of ten specimens, then ten bond strength values of each group (n=10) were employed for statistical analysis using a one-way analysis of variance (ANOVA) and a post-hoc Tukey test, a=0.05. Statistical analysis of the data was performed using a soft ware program; SPSS version 12.

RESULTS

Results of gap formation.

The gap free ratio for the different bonding systems employed (Optibond and XenoIII) are illustrated in Figs. 5and 6 respectively. All specimens of group A; whether Optibond or XenoIII was used, showed marginal gaps. In contrast, neither the specimens of groups B, C, D and E in which Optibond Solo Plus was applied nor specimens of group B in which XenoIII was applied, showed any marginal gaps. However, specimens of groups C, D and E in which XenoIII was applied, showed marginal gaps that were observed more frequently with longer treatment time of NaOCl. Group E demonstrated the highest percentage (88%) of marginal gap among the specimens in which XenoIII was used.

Marginal gap observation

Stereomicroscopic examination showed gap positive when XenoIII was employed after NaOCl treatment (Fig.7-A). Gap negative (gap free) was also observed in groups B, C, D, and E when OptiBond Solo is used (Fig.7-B).

Results of the punch out shear bond strength

Mean shear bond strength and standard deviation are shown in tables 3, 4 and Figures 8, 9. All specimens of group A, regardless the adhesive system employed, showed poor shear bond strengths (0.5 - 0.31MPa). The bond strength values of Optibond for specimens of groups B, C, D and E were 10.1, 8.6, 10.8 and 9.8 MPa, respectively and they were significantly different from that of group A. There were no significant differences in the mean bond strength values among those groups.

The bond strength values of XenoIII for specimens of B and C were not significantly different from each other (p=0.086). However, they were significantly different from D and E (p<0.05).

On the other hand, the specimens treated with XenoIII showed significantly higher bond strength values than those treated with Optibond Solo (p=0.0001).

Table 1. The adhesive systems employed in this study and their composition.

Adhesive system	Composition
Optobond Solo plus (self-priming adhesive system utilizing the total etching technique) **Manufacturer:** Kerr Corp, Orange, Calif.	Ethyl alcohol 20-25% Alkyl dimethacrylate resins 55-60% Barium aluminoborosilicate glass 5-10% Fumed silica (silicon dioxide) 5-10% Sodium hexafluorosilicate 0.5-1%
XenoIII® (Single step self-etching adhesive). **Manufacturer:** DENTSPLY DeTrey GmbH, Germany.	Liquid A -2-hydroxyethyle methacrylate (HEMA) -Purified water -Ethanol -Butylated hyroxy toluene(BHT) -Highly dispersed silicon dioxide Liquid B -Phosphoric acid modified methacrylate resins. -Mono fluoro phosphazene modified polymethacrylate. -Urethane dimethacrylate -Butylated hyroxy toluene(BHT) -Camphorquinone. -Ethyle-4- dimethylaminobenzoate.

Table 2. The application protocol of the adhesive systems.

Adhesive system	Application protocol
Optobond Solo Plus®	-Etch root canal dentin with 15 seconds with 37.5 phosphoric acid etch (kerr gel etchant). Rinse thoroughly for 15 second, then gently blown with compressed air to remove any excess water. Don't desiccate keeping the surface is moist. -Apply OptiBond Solo Plus with a light brushing motion for 15 seconds, then air thin for 3 seconds. -Light cure for 20 seconds. Place composite and light cure.
XenoIII®	-Dispense equal amounts of liquids A and B and mix them for 5 seconds. -Apply the mixture on the root canal dentin surface with a light brushing motion. Leave it for 20 seconds. -Air thinning until there is no more flow. -Light cure for 20 seconds. - Place and light cure composite.

Table 3. Mean and standard deviation of punch out shear strength values of specimens treated with NaOCl and Optibond Solo Plus. Means with different letter exponents are significantly different. (p<0.05)

Groups of Optibond Solo	Mean (MPa)	Standard deviation (±)
A	0.5(a)	0.2
B	10.1(b)	1.7
C	8.5(b)	2.3
D	10.8(b)	3.1
E	9.8(b)	2.2

Table 4. Mean and standard deviation of punch out shear strength values of specimens treated with NaOCl and XenoIII. Means with different letter exponents are significantly different. (p<0.05)

Groups of XenoIII	Mean (MPa)	Standard deviation (+)
A	0.3(a)	0.1
B	6.5(b)	1.9
C	4.8(b)	2.3
D	2.4(c)	1.3
E	1.28(c)	0.7

Fig.1. A specimen is embedded in acrylic resin and mounted on the Milling apparatus.
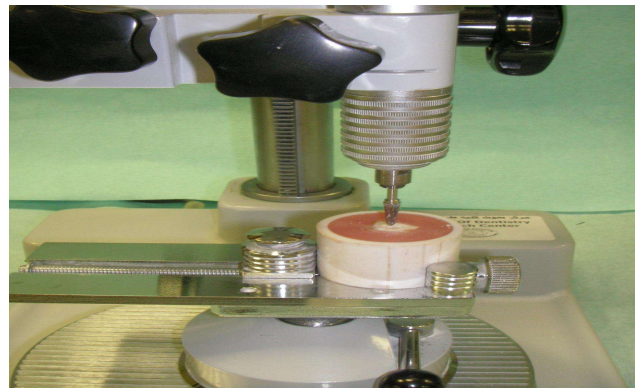

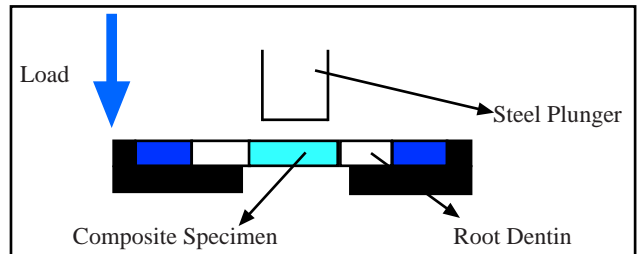**Fig .2. Diagrammatic representation of the set-up of punch out shear test**

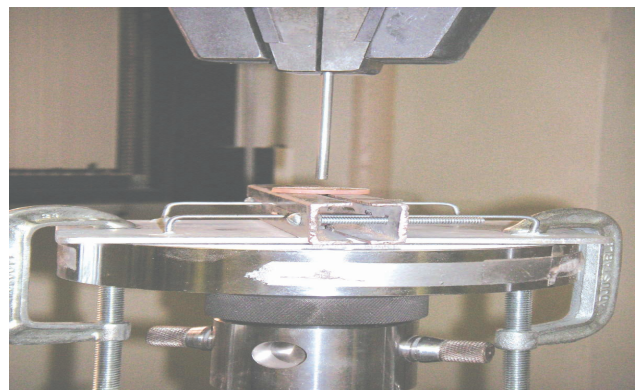Fig.3. A specimen is mounted on the Instron testing machine with the plunger is centered over the specimen.

Fig.7. Steromicroscopic images to identify the gap formation:
A. represents a gap positive as indicated by arrow.
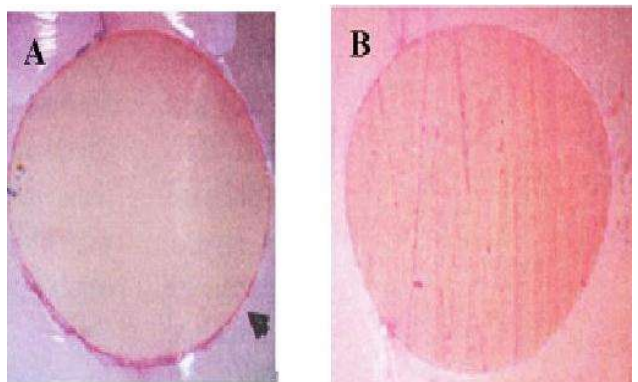B. represents a gap negative (gap free).

Fig.4. Schematic drawing that corresponds to the internal section of the canal walls, Geometric figure for calculation of the cone trunk.
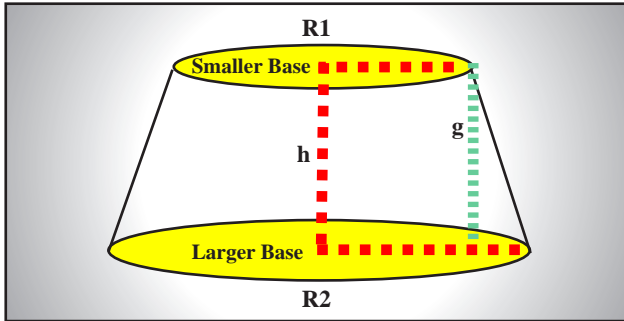

Fig.5. Histogram showing the gap free ratio of specimens treated with NaOCl and Optibond Solo Plus.
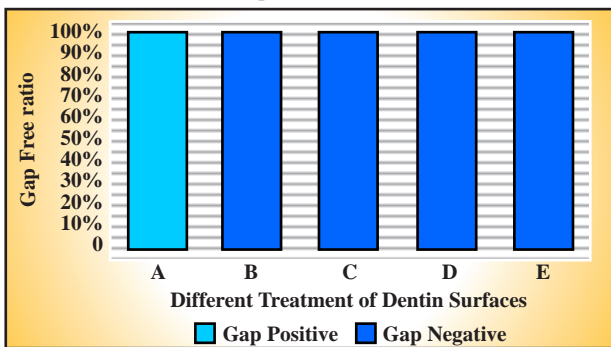

Fig.6. Histogram showing the gap free ratio of specimens treated with NaOCl and XenoIII.
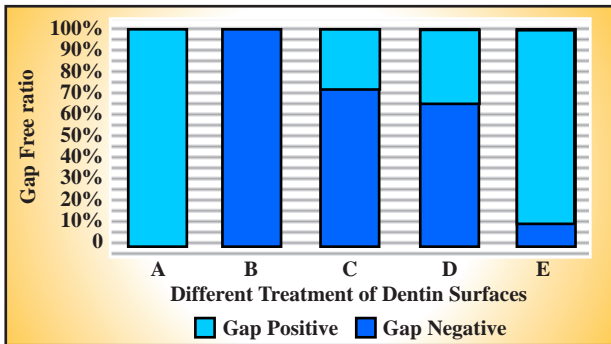

Fig.8. Histogram showing punch out shear bond strength values of different dentin surfaces treated with NaOCl and Optibond Solo.
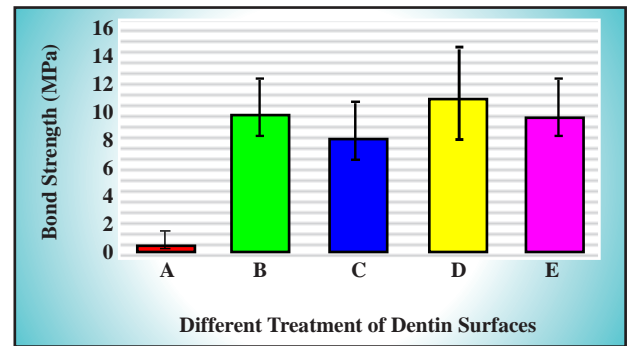

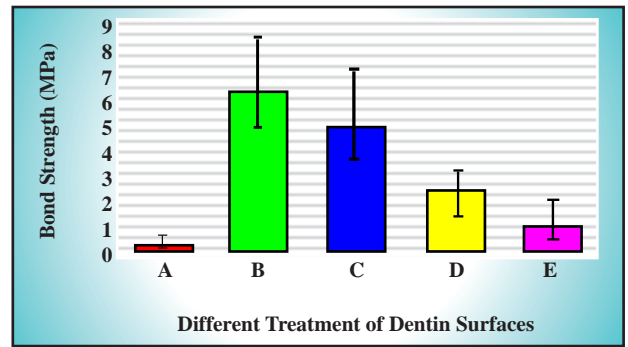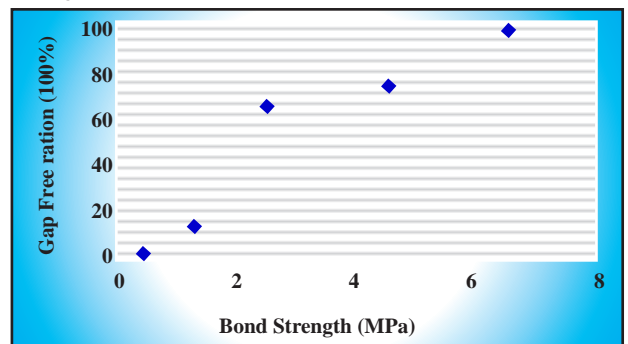Fig.9. Histogram showing punch out shear bond strength values of different dentin surfaces treated with NaOCl and XenoIII.

Fig.10. Relationship between the gap formation ratio and bond strength of XenoIII at different treatmants with NaOCl.

DISCUSSION

There are only a few studies[17,24] that evaluated resin bonding efficiency to the root canal dentin or deep coronal dentin. In these studies, flat dentin surfaces were sectioned along the root and then cylindrical composite resin specimens were adhered to these surfaces for conventional shear bond strength evaluation. In the present study, the cavity was prepared inside the root canal dentin wall and the adhesive systems were applied from the pulpal side. In addition, almost all directions of the dentinal tubules were perpendicular to the cavity wall. The convergence angle of the prepared cavity was 5°, which was similar to that of a tapered fissure-bur for dowel space preparation. Therefore, the cavity preparation used was considered to be typical for composite resin restoration after root canal treatment. Perfect marginal adaptation of a composite resin to dentin is essential to avoid marginal discoloration and leakage. There are several methods to evaluate marginal adaptation, such as microleakage test and wall-to-wall gap observation in ISO/TR11405: 1994.[25] In the present study, the observation method of the contraction gap similar to ISO/TR11405: 1994 was employed. The tensile bond or shear bond test is generally employed to evaluate the resin bonding strength to dentin. However, the punch out shear bond test has been suggested to evaluate the shear strength of composites.[26]

The experimental set-up of this study, could measure both marginal adaptation and shear bond strength using the same specimen. Moreover, it was possible to apply the load perpendicular to the cavity wall which consisted of root canal wall dentin. Therefore, this method could be employed to evaluate adhesion of luting materials for dowel post systems to the root canal wall. However, there are a few disadvantages in this method according to Nomoto et al.[27] For example, the results are influenced by the modulus of elasticity of the filling materials used; punch out bond strength is increased when a filling material with a low elastic modulus is employed because the filling material deforms during loading, which interferes with the pushout from the cavity.[26] Therefore, truncated cone cavities were employed in the present study to avoid interference caused by the filled material's deformation. The configuration of cavity preparation is an influential factor for wall-to-wall gap formation.

Recently, the "c-factor" was introduced to determine the cavity configuration effect on resin bond strength to dentin.[10,28] The apical side of the prepared cavity was faced with gutta-percha which did not adhere to the composite resin in the present study. Therefore, the c-factor of the prepared cavity was calculated to be approximately 1.25; the value was similar to a class IV or III cavity, which might be favorable for resin bonding to dentin.[28]

The dentin bonding systems used in the present study represent the total etching adhesive (optibond SoloPlus) and self-etching adhesive (XenoIII). In the total etching system, the etchant removes the smear layer and demineralizes dentin, and the adhesive resin penetrates the etched dentin. However, the acid monomer of the self-etching adhesive mildly demineralizes dentin so that it does not remove smear plugs completely. At the same time, the primer component modifies the demineralized dentin and the bonding resin infiltrates the primed dentin.

Several studies reported on the marginal adaptation and bond strengths of total etching adhesive systems (Single Bond, 3M).[17,29-34] Bond strengths obtained from these studies were reported to be in a range of 12.8-20.1 MPa. While in the present study, bond strength was lower than that obtained in previous studies, because those studies applied bonding systems to flat dentin surfaces and c-factor was smaller than that of the present study.

There have been several studies reporting the effects of endodontic irrigants on resin bonding systems. In particular, the effects of NaOCl have been thoroughly discussed but not clearly confirmed. The loss of physical properties of NaOCl-treated dentin could not be reversed after resin infiltration. This may be due to a lack of proper interaction of the adhesive agent with the altered dentin substrate. Several studies reported a decrease in bond strength of Single Bond to NaOCl-treated dentin.[35-36]

In this study the shear bond strength was reduced with increasing the application time for sodium hypochlorite. This may be due to incomplete polymerization of the self-etching adhesive and to reduced mechanical properties of the infiltrated resin. The presence of reactive residual free-radicals as a result of the oxidizing action of NaOCl, may compete with the propagating vinyl free-radicals generated during light-activation of the adhesive, resulting in premature chain termination and incomplete polymerization.[18,24]

This is the most likely phenomenon that occurred in this study; the inability of the infiltrating resin to recover the strength of the NaOCl-treated dentin was probably caused by its incomplete polymerization reaction as results of presence of residual NaOCl and the presence of oxygen may interfere with the polymerization of the bonding resin.

The bond strength of resin following NaOCl treatment before etching decreased when a MMA-TBB resin system was employed.[37] The decreased bond strength is improved when an ascorbic acid or a sodium thiosulfate solution is applied after NaOCl treatment; these solutions remove NaOCl by the oxidation-reduction reaction.[18]

Nikaido et al.[16] evaluated the bonding strength at the buccal dentin surface after NaOCl treatment on root canal wall dentin ; the bonding strength of a total etching adhesive (single Bond) was significantly decreased after NaOCl treatment, while that of a self-etching primer system did not change. In the present study, the bonding strength of XenoIII (a self-etching adhesive) decreased following NaOCl treatment whereas that of Optibond Solo (a total etching adhesive) did not change. The results of this study were in contradiction to the results of Nakaido et.al. Therefore, the effect of NaOCl treatment on bonding strength may depend on the location of NaOCl application. XenoIII might not be effective for removing degenerated dentin and residual NaOCl, while the etchant of Optibond Solo might be strong enough to remove both. Moreover, the effects of NaOCl might be different between superficial dentin and root canal wall dentin. These may explain the discrepancy between the present findings and those previously reported.

There are many studies that evaluated the relationship between marginal adaptation and bond strength. Some studies have suggested no correlation between bond strength and marginal adaptation,[38] while others suggested that higher tensile bond strength meant lower dye microleakage.[32] In the present study, the punch out bond strength did not always show a smaller value even when marginal gaps were observed. The scatter diagram showing the relationship between the punch out bond strength and gap free ratio of XenoIII is shown in Fig.10. The gap free ratio increased to 100% with the bond strength up to 6.5 MPa, it means no gap formation was recorded. On the other hand, the bonding strength of Optibond, more than 6.5MPa, showed 100% gap free ratio. These findings suggest that the marginal gap may not be produced when the bond strength is greater than a certain value; the bond strength of 6.5 MPa was considered to be the threshold for gap formation of the composite resin and the adhesive systems employed in this study.

However, the findings of this study were obtained after a relatively short water storage period without thermal stress. It is necessary to evaluate the changes of marginal adaptation and bond strength using a longer water storage period under thermal stress.

CONCLUSIONS

1 The experimental design for examining the marginal adaptation and punch out bond strength simultaneously was considered to be effective for evaluating resin bonding efficiency to root canal dentin.

2 The null hypothesis was rejected, that was the treatment of root canal with NaOCl would not compromise dentin bonding.

3 NaOCl interferes with the bonding ability of XenoIII but does not affect that of Optibond Solo.

4 The marginal gap may not be produced when the bond strength is greater than a certain value; the bond strength of 6.5 MPa was considered to be the threshold for gap formation of composite resin and root canal dentin.

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