Regional bond strength of dentin bonding systems to pulp chamber dentin

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INTRODUCTION

The development of new dentin bonding systems has brought about great improvements in adhesive dentistry and the use of resin composite has become widely accepted for treatment of both anterior and posterior teeth. Also resin composites used as core materials after endodontic treatment are becoming more and more popular in the clinical practice of dentistry because of their esthetics, ability to bond to tooth structure, strength, and the fact that they allow immediate continuation of a crown preparation. Good adhesion between the restorative material and the cavity wall results in good marginal sealing, less microleakage, and longevity of the restoration11.

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Endodontic treatment has become a routine procedure for treating and retaining nonvital teeth. Endodontic treatment consists of removing all contents of the root canal system before and during cleaning and shaping. Successful cleaning entails the use of instruments to mechanically remove dentin, irrigant to flush loosened debris away, and chemicals to dissolve contaminants from inaccessible regions.

Sodium hypochlorite and hydrogen peroxide are common endodontic irrigants that are used for the debridement and deproteinization of mechanically prepared, smear-layer-covered radicular dentin. Sodium hypochlorite is also frequently used for chemomechanical caries removal and the arrest of hemorrhage in pulpal exposures before bonding to coronal dentin occurs. The use of these irrigants provides gross debridement, lubrication, destruction of microbes, and dissolution of tissues.

Recent studies showed that bond strength of some adhesives was compromised by the use of these reagents on root and crown dentin, as well as enamel. Contamination of dentin with blood or other body fluids can also be detrimental to bond strength with dental adhesives.

The incomplete removal of the partially denatured or destabilized collagen matrix has been proposed as a possible reason for compromised bond strength in sodium hypochlorite-treated, acid-etched dentin. Sodium hypochlorite, apart from being an effective deproteinizing agent, is similar to hydrogen peroxide in that it is also a potent biological oxidant. Sodium hypochlorite breaks down to sodium chloride and oxygen. This oxygen causes strong inhibition of the interfacial polymerization of resin bonding materials. After the chemical irrigation of the root canal, the residual chemical irrigants and their products are likely to diffuse into the dentin along the dentinal tubules, which must result in decrease of bond strength.

The tooth structure that remains after endodontic treatment may be undermined and weakened by caries, fracture, tooth preparation, and restoration. Endodontic manipulation further removes important intracoronal and intraradicular dentin. Finally, the endodontic treatment changes the actual composition of the remaining tooth structure.

The major changes in the endodontically treated tooth include loss of tooth structure, altered physical and esthetic characteristics of the residual tooth. The decreased strength evaluated in endodontically treated teeth is primarily due to the loss of coronal tooth structure and is not a direct result of the endodontic treatment. Endodontic access into the pulp chamber destroys the structural integrity provided by the coronal dentin of the pulpal roof and allows greater flexing of the tooth under function. However, the tooth structure remaining after endodontic treatment also exhibits irreversibly altered physical characteristics. Changes in collagen cross-linking and dehydration of the dentin result in a 14% reduction in strength of endodontically treated molars. The internal moisture loss has been shown to average approximately 9%.

Dentin is not a uniform tissue but differs from each region. With age, dentin formation continues slowly, and the regular secondary dentin is laid down at the pulp end of the primary dentin. In response to noxious stimuli, the odontoblasts may evacuate the tubules, giving rise to the so-called dead tract, and/or seal off the tubules at their pulpal ends with irregular secondary dentin, or form sclerotic dentin. Irregular secondary dentin has many fewer tubules with irregular orientation, than primary dentin.

Associated with physiological aging, especially in root dentin, the dentinal tubules become completely occluded by mineral in a process similar to that of peritubular dentin formation. Fogel et al. showed that the permeability of root dentin is much lower than that of coronal dentin. The number of dentinal tubules per unit area is less for radicular dentin, meaning that the area of intertubular dentin available for bonding is greater in the root than the crown. The structure of pulp chamber floor is complicated, including primary dentin, and regular and irregular secondary dentin.

Restorations for endodontically treated teeth should be designed to replace the missing tooth structure and to protect the remaining tooth structure from fracture. Not every endodontically
treated tooth needs a crown or a dowel. Recently, the opportunity for the restoration of nonvital teeth with resin composite has increased based on the development of better dentin bonding systems. A resin composite enables a nonvital tooth to be restored by only replacement of the missing tooth structure, because adhesive restoration can reinforce remaining tooth structure\textsuperscript{22-23}.

Adhesive restoration for endodontically treated teeth offers many advantages over the use of traditional, nonadhesive materials. For instance bonded resins permit transmission of functional stresses across the bonded interface to the tooth, with the potential to reinforce weakened tooth structure\textsuperscript{24}. Using adhesive materials properly can reduce microleakage of interface between these materials and tooth structure. Application of adhesive to acid-etched dentin creates an acid-resistant, resin-infiltrated collagen layer, the so-called hybrid layer that not only retains composites to dentin, but also can seal dentin from oral fluids\textsuperscript{25}.

Recently, Sano et al\textsuperscript{26} developed micro-tensile bond strength test, permitting the measurement of small bonded areas as small as 1 mm\textsuperscript{2}. As this new method permits measurement on small areas, it can be used to compare regional bond strengths with different surface of the pulp chamber.

The purpose of this study is to evaluate the regional bond strengths of three dentin bonding systems to pulp chamber dentin of endodontically treated teeth and to evaluate the effect of NaOCl pretreatment on resin–dentin bonding interface.

\section{MATERIALS AND METHODS}

Forty-five caries-free human molars extracted for the periodontal reasons were used in this study. The teeth were stored frozen after extraction until use. Initially, the teeth were embedded in epoxy resin using acrylic ring. Then, teeth were divided into three groups of fifteen for each control and experimental groups. Three dentin bonding systems and one resin composite were used in this study and their components, manufacturers were listed in Table 1.

\subsection{Specimen preparation}

(1) Control group — Intact dentin group
The teeth were sectioned to remove occlusal

| Table 1. Materials used in this study |
|--------------------------------------|
| Material (code) | Component | Composition | Manufacturer |
|-----------------|-----------|-------------|--------------|
| Scotchbond Multi-Purpose (SM) | Etchant | 35\% Phosphoric acid | 3M Dental Products, St. Paul, MN, USA |
| | Primer | HEMA, water, Polyalkenoic acid copolymer | |
| | Adhesive | HEMA, Bis-GMA | |
| Single Bond (SB) | Etchant | 35\% Phosphoric acid | 3M Dental Products, St. Paul, MN, USA |
| | Adhesive | HEMA, Bis-GMA, Polyalkenoic acid copolymer, ethanol, water | |
| Clearfil SE Bond (SE) | Primer | MDP, HEMA, water | Kuraray Co., Osaka, Japan |
| | Adhesive | MDP, HEMA, dimethacrylate, microfiller | |
| Z-100 Restorative composite | Resin | Zirconia/silica filler | 3M Dental Products, |
| | | Bis-GMA, UDMA | St. Paul, MN, USA |

Bis-GMA = Bisphenol-A glycidyl methacrylate
HEMA = Hydroxyethylmethacrylate
MDP = Methacryloyloxydecyl dihydrogen phosphate
enamel using a Low speed diamond saw (ISOMET; Buhler, USA) and exposed dentin surface was ground with #600-grit SiC paper serially under a stream of running water. Three dentin bonding systems, Scotchbond Multi-Purpose (3M, USA), Single Bond (3M, USA), Clearfil SE Bond (Kuraray, Japan), were applied according to manufacturer’s instruction (Table 2). Then, teflon mold (diameter: 6mm, height: 2mm) was placed on bonding area and filled with resin composite (Z-100: 3M, USA) and light-cured (Spectrum 800: Dentsply, USA) for 40 sec.

(2) Experimental group
Initially, the access cavity preparation was performed using high-speed diamond point under copious water spray. Following this, pulp tissue in pulp chamber was carefully removed using endodontic file. The teeth were then stored in 5% NaOCl for 1 hour.

## Table 2. Instruction for dentin bonding systems

| Product          | Instruction                        |
|------------------|------------------------------------|
| Scotchbond Multi-Purpose | Etching 15 sec, priming, air dry 5 sec |
| Single Bond      | Adhesive (two coat), air dry 5 sec  |
| Clearfil SE Bond | Primer 20 sec, air dry Adhesive, light-cure 10 sec |

1) Axial wall group
The teeth were sectioned mesio-distally parallel to the long axis of teeth using Low speed diamond saw. Then, three dentin bonding systems were applied to axial wall area in pulp chamber according to manufacturer’s instruction and Z-100 was filled in pulp chamber and light-cured for 40 sec.

2) Pulpal floor group
The teeth were sectioned at pulpal floor level perpendicular to the long axis of teeth and ground with #600-grit SiC paper serially. Three dentin bonding systems were applied according to manufacturer’s instruction. Then, teflon mold was placed on the cured bonding resin and filled with Z-100 and light-cured for 40 sec.

2. Micro-tensile bond strength test
All restored specimens were stored in distilled water at 37°C for 24 hours. The teeth were serially sectioned into slice of mean thickness 0.7 mm perpendicular to the bonded surface using a Low speed diamond saw under copious water supply. These specimens were then trimmed into an hour-glass shape to give a bonded surface of 1 mm² using a high speed diamond point (#104: Shofu, Japan). The trimmed specimens were attached to testing zig with cyanoacrylate adhesive(Zapit: MDS Products Co., USA), then subjected to tensile forces in a universal testing machine(EZ Test, Shimadzu, Japan) at a cross head speed 1 mm/min.

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Fig. 1. Specimen preparation for micro-tensile bond strength test (Intact dentin group, Pulpal floor group)  
Fig. 2. Specimen preparation for micro-tensile bond strength test (Axial wall group)
3. SEM evaluation

For the evaluation of the morphology at the resin-dentin interface, specimens used for hybrid layer observation were bonded with each system in the same manner as for the micro-tensile bond test. One day later, the teeth were sectioned perpendicular to bonding surface and then embedded in epoxy resin. Then the sectioned surfaces were serially ground to #2000-grit SiC papers, and highly polished with a diamond paste. The specimens were subjected to 10% phosphoric acid treatment for 3~5 sec. Then specimens were rinsed with water for 15 sec and treated with 5% sodium hypochlorite for 5 min. After being extensively rinsed with water, the treated specimens were air dried, gold-sputter-coated and examined in SEM (S-2300; Hitachi Co., Japan).

4. Statistical analysis

The maximum tensile force was divided by the area of the specimen and the measured micro-

tensile bond strength values were analysed using ANOVA/ Newman-Keuls multiple comparisons test at a significance level of 0.05.

III. RESULTS

1. Micro-tensile bond strength

The micro-tensile bond strengths of Scotchbond Multi-Purpose (SM), Single Bond (SB) and Clearfil SE Bond (SE) with intact dentin, axial wall and pulpal floor are shown in Table 3.

As shown in Table 3, the micro-tensile bond strength of all dentin bonding systems were decreased in order of control group, axial wall group, pulpal floor group. In control group, SM and SB showed significantly higher bond strength than SE (p<0.05). However, in axial wall and pulpal floor groups, there were no significant difference between dentin bonding systems.

The micro-tensile bond strengths of SM and SB were much higher in control group than that of axial wall and pulpal floor group (p<0.05).

For SE, control group showed significantly higher bond strength than axial wall and pulpal floor group. Also pulpal floor group showed significantly lower bond strength than control and axial wall group (p<0.05).

The micro-tensile bond strengths according to locations of dentin and to dentin bonding systems are showed in Fig. 3 and 4.

| Table 3. Micro-tensile bond strength of each bonding system to dentin (MPa±S.D, n=10) |
|---|---|---|---|
| Group | Control | Axial | Pulpal |
| SM  | 36.71±5.36 | 24.64±9.73 | 21.73±8.12 |
| SB  | 34.69±6.31 | 23.85±6.86 | 20.54±6.80 |
| SE  | 28.04±6.23 | 22.64±5.71 | 19.43±5.39 |

Fig. 3. Micro-tensile bond strengths according to location of dentin

Fig. 4. Micro-tensile bond strengths according to location of dentin

Regional bond strength of dentin bonding systems to pulp chamber dentin
2. SEM Evaluation

In SEM observation, there were several notable regional differences in dentin structure. The tubule density was much lower on the floor of the pulp chamber compared with the intact dentin. For axial wall and pulpal floor group, which were treated with 5% NaOCl, predentin matrix was removed, leaving a smoother, mineralized matrix for bonding. This lead to smoother interfaces. Most of the bond failures occurred at the top of the hybrid layer. For SM, the hybrid layer thickness ranged from between 4-5 μm. Resin tags were

Fig. 5. SEM photograph of the adhesive interface of SM-control group (×2000)

Fig. 6. SEM photograph of the adhesive interface of SM-axial wall group (×2000)

Fig. 7. SEM photograph of the adhesive interface of SM-pulpal floor group (×2000)

Fig. 8. SEM photograph of the adhesive interface of SB-control group (×2000)

Fig. 9. SEM photograph of the adhesive interface of SB-axial wall group (×2000)
clearly observed with the typical funnel shape at
the top of the tubules and more than 10 μm in
length. SB exhibited similar pattern to SM but
the thickness of the hybrid layer was thin (2–3
μm) and the length of resin tag was relatively
short (5–7 μm). For SE, the thickness of the
hybrid layer was measured between 4–5 μm. Resin
tags were not observed in the pulpal floor dentin.
In other regions where resin tags were present,
they were thin and poorly formed.

IV. DISCUSSION

Previous studies reported that endodontically
treated teeth are not reinforced by a full coverage
crown combined with post and core systems.29-30)

Those studies stated that preservation of the
tooth structure is an important factor to prolong
the longevity of the tooth. In addition, minimizing
the amount of tooth structure loss is reported to
be essential for a favorable prognosis.16) With
development of adhesive dentistry, the weakened
tooth structure can be reinforced by the use of
resin bonding system after endodontic treatment.
However, both the composition and morphology
of pulp chamber dentin are different from those of
intact dentin. At the surface of the dentin, or the
dentino-enamel junction, dentinal tubules range
between 15,000 and 20,000/mm². At the pulpal
surface, the number of dentinal tubules increases
three fold to 45,000 to 60,000/mm² and the
tubule diameter increases. Dentin permeability is
greatest on thin axial surfaces, particularly mesial surfaces\(^{16}\). Also, with aging or in response to noxious stimuli, secondary dentin is laid down at the pulpal end of the primary dentin. This secondary dentin has fewer dentinal tubules, irregular orientation, and lower permeability than primary dentin. Especially, most of pulpal floor composed with calcified secondary dentin.

In addition, pulp chamber dentin might be affected by root canal irrigants and disinfectants during endodontic procedure. NaOCl is one of the most common root canal irrigants used for debridement, lubrication, destruction of microbes and dissolution of organic tissues\(^{2}\). Nikaido et al\(^{7}\) reported that NaOCl treatment adversely affected the bond strengths to dentin. In contrast, others reported that NaOCl treatment improved the adhesion of the bonding system to dentin when using phosphoric acid\(^{31}\).

In this study, the micro-tensile testing method was used for regional bond strength of pulp chamber dentin. Since, each region of pulp chamber dentin is too small to permit conventional bond test and micro-tensile testing method has been shown that the bonded interface of small specimens distributes stress better, which can result in more consistent adhesive failures and higher apparent bond strengths. It also allows for more specimens to be generated from the same tooth. One of the disadvantage of the test is that small bond strengths are difficult to measure because specimens can be broken easily during preparation\(^ {32}\).

The dentin bonding systems used in this study represent the conventional bonding system (SM), self-priming system (SB) and self-etching system (SE). In conventional and self-priming system, the etchant removes the smear layer and demineralizes dentin, and the adhesive resin penetrate the etched dentin. However, the acid component of the self-etching primer 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.

The micro-tensile bond strength of pulp chamber dentin (axial wall, pulpal floor) was lower than that of intact dentin (control) in all dentin bonding systems. This result suggests that endodontic treatment which use chemical irrigant such as NaOCl interfere the adhesion of bonding system to dentin. It is thought that residual NaOCl may interfere with polymerization of the bonding resin due to oxygen generation. For SM and SB, micro-tensile bond strength was decreased so much in pulp chamber dentin, but SE showed relatively gradual decreation though there were no significant difference between dentin bonding systems in pulp chamber dentin. Several factors could explain this difference between wet bonding system (SM, SB) and self-etching system (SE). The self-etching primer of SE might not be effective for removing degenerated dentin and residual NaOCl, while the etchant of SM, SB might be strong enough to remove both. The remained smear layer after self-etching primer application might prevent oxygen from penetrating through dentinal tubules from the pulp chamber dentin, while the removal of the smear layer by the etchant treatment of SM, SB might allow oxygen penetration. In highly calcified pulpal floor dentin, self-etching primer might not etch mineral enough to infiltrate by monomers to form a hybrid layer. Self-etching primer could potentially eliminate the risk of overetching dentin. Overetching leaves a layer of demineralized dentin below the hybrid layer, leading to long-term weakening of the dentin bond and subsequent leakage\(^ {33}\). Nikaido et al\(^{7}\) have shown that bond strength of self-priming system was significantly decreased, while self-etching system did not affected by NaOCl treatment. Ishizuka et al\(^{34}\), on the other hand, found that the bond strength of self-etching system decreased following NaOCl treatment whereas that of wet bonding system did not change. In this study, although bond strengths of all dentin bonding systems were affected by NaOCl, self-etching system was less affected by NaOCl treatment. It is thought that as the self-etching primer acts on the dentin surface by modifying the smear layer, and partially dissolved smear plugs may remain within the tubules, low-
ering the dentinal permeability. Therefore residual chemical irrigants and their products in dentinal tubules are main reason that affect the penetration of resin into the dentin structure and/or the polymerization of the monomer in the demineralized dentin.

In SEM observation, the thickness of hybrid layer or the length of resin tag were not affect the bond strength, supporting previous study that there is no correlation between bond strength and hybrid layer thickness. But smoother bonding interfaces due to use of NaOCl as well as lower dentinal tubule density were thought another reason of lower bond strength in pulp chamber dentin. Although the bond strength of SE was slightly lower than that of SM, SB in pulp chamber dentin, the intent of use of adhesive resins inside the pulp chamber is to seal the root canal to prevent microleakage of oral microorganisms and their products, then high bond strength are not required, because the correlation between microleakage and bond strength is not high. Rather the use of adhesive system that is simple, easily retreatable, and technique-insensitive would be a good choice.

In addition, we tried to reduce the C-factor (cavity configuration factor) in order to exclude the effect of C-factor that may affect the bond strength. For instance, in control and pulpal floor group we made flat dentin surface, also dentin bonding systems were applied only to the axial wall of pulp chamber in axial wall group. However, when the pulp chamber is restored with resin composite, special care must be taken to reduce C-factor which may cause microleakage.

In this study, endodontic procedure prove to have an adverse effect on bond strengths of composite. Wet bonding system was more influenced by chemical irrigant than self-etching system even though bond strengths of wet bonding system were slightly higher. Then, for the recovery of bond strength in endodontically treated teeth, the use of anti-oxidant such as sodium ascorbate before resin bonding may be considerable.

V. CONCLUSION

In restoring endodontically treated teeth, treatment goals must be based upon a multitude of factors that include occlusion, patient's function, tooth position, periodontal status, prosthetic needs, amount of remaining tooth structure, and root morphology. Recent development of adhesive dentistry enables a non-vital tooth to be restored with minimal intervention.

This study was designed to evaluate the effect of endodontic treatment on bond strength to pulp chamber dentin. From the results of this study, we can conclude as follows:
1. The micro-tensile bond strengths of all dentin bonding systems were decreased in order of control (intact dentin) group, axial wall group, and pulpal floor group.
2. In control group, SM and SB showed significantly higher bond strengths than SE (p(0.05).
3. SM and SB showed significantly lower bond strengths in axial wall and pulpal floor group when compared with control group (p(0.05). But SE showed significantly lower bond strength only in pulpal floor group than control group (p(0.05).
4. In axial wall and pulpal floor group, there were no significant differences between dentin bonding systems.
5. In SEM observation, the tubule density was much lower on the floor of the pulp chamber compared with the intact dentin. For axial wall and pulpal floor group, which were treated with 5% NaOCl, the smoother bonding interface was shown. There was no correlation between the bond strength and the thickness of hybrid layer.

This study suggests that the procedure of endodontic treatment using chemical irrigants can adversely affect the adhesion to dentin, but self-etching system is less affected than wet bonding system which needs total etching procedure. Therefore, proper selection of dentin bonding system is important in restoring endodontically treated teeth with resin composite.
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