



# Smear Layer-Deproteinization: Improving the Adhesion of Self-Etch Adhesive Systems to Caries-Affected Dentin

Keiichi Hosaka<sup>1</sup> · Taweesak Prasansuttiorn<sup>2</sup> · Ornnicha Thanatvarakorn<sup>3</sup> · Sitthikorn Kunawarote<sup>2</sup> · Masahiro Takahashi<sup>1</sup> · Richard M. Foxton<sup>4</sup> · Junji Tagami<sup>1</sup> · Masatoshi Nakajima<sup>1</sup>

© Springer International Publishing AG, part of Springer Nature 2018

## Abstract

**Purpose of review** This paper reviews a new method of dentin surface modification, smear layer-deproteinization for self-etch adhesive systems, particularly in relation to improving the adhesion to caries-affected dentin.

**Recent Findings** Remnants of smear debris, which forms hybridized smear layer with self-etch adhesives, can prevent monomer infiltration and interfere with the chemical interaction of adhesive monomers and the underlying dentin. The hybridized smear layer weakens the physical and chemical properties of the resin-dentin hybridized complex both immediately and over time. Smear layer-deproteinization with NaOCl and HOCl solutions can improve the quality of resin-dentin interface of self-etch adhesives through elimination of the hybridized smear layer, development of monomer infiltration, and enhancement of the chemical interaction of adhesive monomers with hydroxyapatite due to an increase in the mineral/organic ratio on the dentin surface. These positive effects are influenced by the types of oxidizing solution and their application time and also depend upon the adhesive materials used because compromising effects of residual oxidized-byproducts at the dentin surface on the polymerization behavior of the adhesives are different between the materials. However, applying antioxidant/reducing agents can eliminate this problem.

**Summary** Smear layer-deproteinization is more effective for improving the bonding efficacy of self-etch adhesives to caries-affected dentin than normal dentin because caries-affected dentin produces a thicker organic-rich smear layer. Smear layer-deproteinization with HOCl solution, which has a rapid and broad-spectrum antimicrobial activity with less irritating and sensitizing properties, along with the subsequent application of antioxidant/reducing agents could enhance the longevity of composite restoration with self-etch adhesives.

**Keywords** Self-etch adhesive · Smear layer · Smear layer-deproteinization · Oxidizing solution · Antioxidant/reducing agent · Caries-affected dentin

This article is part of the Topical Collection on *Dental Restorative Materials*

✉ Masatoshi Nakajima  
nakajima.ope@tmd.ac.jp

<sup>1</sup> Cariology and Operative Dentistry, Graduate School of Medical and Dental Sciences, Tokyo Medical and Dental University, 1-5-45 Yushima, Bunkyo-ku, Tokyo 113-8549, Japan

<sup>2</sup> Department of Restorative Dentistry and Periodontology, Faculty of Dentistry, Chiangmai University, T. Suthep, A. Muang, Chiang Mai 50200, Thailand

<sup>3</sup> Faculty of Dentistry, Bangkokthonburi University, 16/10 Leabklongtaweewatana Rd., Khet Taweewatana, Bangkok 10170, Thailand

<sup>4</sup> Restorative Dentistry, King's College London Dental Institute at Guy's, King's and St Thomas' Hospitals, King's College London, Floor 21, Guy's Hospital, London SE1-9RT, UK

## Introduction

The dentin surface is covered with a smear layer following cavity preparation with rotary instruments whereby the dentinal tubules are occluded with smear plugs. This smear layer does not have a physiological or morphological continuous connection to the underlying dentin. Moreover, the smear layer can prevent adhesive monomer infiltration and interfere with the interactions of functional monomers with hydroxyapatite derived from the underlying dentin. Therefore, in order to achieve ideal adhesion to the dentin substrate, this dentin smear layer should be removed.

Clinically, self-etch adhesive systems are widely used because of superior bonding performance to enamel and dentin, simplified bonding procedures, and reduced postoperative

sensitivity. Additionally, self-etch adhesive systems exhibit reduced nanoleakage formation compared with etch and rinse adhesive systems using phosphoric acid etching, because they can simultaneously demineralize the dentin surface, infiltrate adhesive monomers, and form a thin hybrid layer (approximately 1  $\mu\text{m}$  thick). An adhesive interface formed with reduced nanoleakage formation is expected to provide stable bonding even in the long run.

On the other hand, self-etch adhesive systems cannot completely remove the smear layer due to their mild acidity. Surface preparation methods affect the dentin smear layer characteristics (such as thickness, particle size, density), which affect the bonding performance and change the dentin surface conditions with residual amount of smear debris after treatment with self-etch adhesive systems [1], depending on the pH of the adhesives [2]. Moreover, the smear layer characteristics also affect the long-term bonding stability of self-etch adhesives [3]. Tay and Pashley demonstrated that a resin-impregnated smear layer (so-called hybridized smear layer) is formed on the authentic hybrid layer by incorporating remnants of smear debris [4]. Remnants of smear debris could act as a selective barrier for monomer infiltration into the underlying dentin and interfere with the interaction of self-etch systems and the underlying dentin [5, 6]. A hybridized smear layer would weaken the physical and chemical properties of the resin-dentin hybridized complex. Elimination of the hybridized smear layer could lead to a higher quality hybridized complex between dentin and self-etch adhesive systems.

The dentin smear layer is composed of disorganized collagen debris-binding mineral particles [7]. The disorganized collagen within the smear layer is not denatured, but forms a gelatinous matrix around the mineral particles and cannot be easily removed even with phosphoric acid etching [7]. Self-etch adhesive systems are mildly acidic when compared with phosphoric acid and can therefore dissolve mineral components and partially remove the mineral phase of the smear layer. However, they cannot dissolve organic components and the organic phase within the smear layer therefore remains on the dentin surface. Remnants of the organic phase in the smear layer might be in essence, hybridized smear layer above the authentic hybrid layer formed with self-etch adhesives. Therefore, deproteinization of the smear layer prior to application of self-etch adhesives would lead to elimination of the hybridized smear layer and facilitate adhesive monomer infiltration into the dentin substrate [8••, 9••]. Additionally, deproteinization of the smear layer would lead to an increase in the mineral/organic ratio on the smear layer-covered dentin surface and a decrease of surface water content due to removal of hydrated organic phase [8••, 10–12], which would be advantageous for chemical interactions of adhesive monomers with hydroxyapatite in dentin [11, 12].

## Smear Layer-Deproteinization with NaOCl and HOCl Solutions

A 2.5–6% NaOCl solution is widely used as an endodontic antiseptic and irrigation solution, due to its debriding, deproteinizing, and antimicrobial properties. However, it is an irritant and is cytotoxic because of its extreme alkalinity (pH 10.8–13.2). NaOCl dissociates into  $\text{Na}^+$  and  $\text{OCl}^-$  ions and establishes HOCl in water. The proportion of HOCl and  $\text{OCl}^-$  ions in the solution depends on the pH [13]. That is, above pH 9, almost all HOCl ionizes to  $\text{OCl}^-$ , and at a pH of approximately 6, the majority of the chlorine is in the form of HOCl [13]. Therefore, in NaOCl solution, almost all the chlorine is present in the  $\text{OCl}^-$  form. The  $\text{OCl}^-$  ion is the determining factor for proteolytic activity, because the  $\text{OCl}^-$  ion gives rise to protein chlorination and chloramines, which are then broken down into nitrogen-centered radicals and induce the fragmentation of proteins [14]. NaOCl treatment for 20, 40, and 120 s can increase the mineral to matrix ratio in smear-layer covered dentin by dissolution of the gelatin-converted collagen part of the smear layer [10].

HOCl is a major inorganic bactericidal compound, which provides innate immunity. Owing to its strong chlorinating and oxidizing actions, HOCl exhibits a rapid reaction in a broad range of microorganisms and organic materials [13, 15], and is considered to be 20–300 times more effective for antimicrobial ability than  $\text{OCl}^-$  [16]. Additionally, in agricultural science, HOCl leaves fewer residues on the treated surface than NaOCl. It has been stated that the high reactivity of NaOCl to amino acids makes washing it away difficult even after several rinses with water [17]. NaOCl solution leaves significantly higher chlorine residues on dip-treated surfaces (spinach and lettuce leaves) than HOCl [18]. HOCl solution, such as Comfosy (Haccpper Advance), which is developed at pHs of 7.0–6.4 by neutralizing NaOCl with HCl, has outstanding properties because not only does it have immediate and highly effective antimicrobial and deproteinizing properties even at lower chlorine concentrations, but is also biocompatible and has low cytotoxicity.

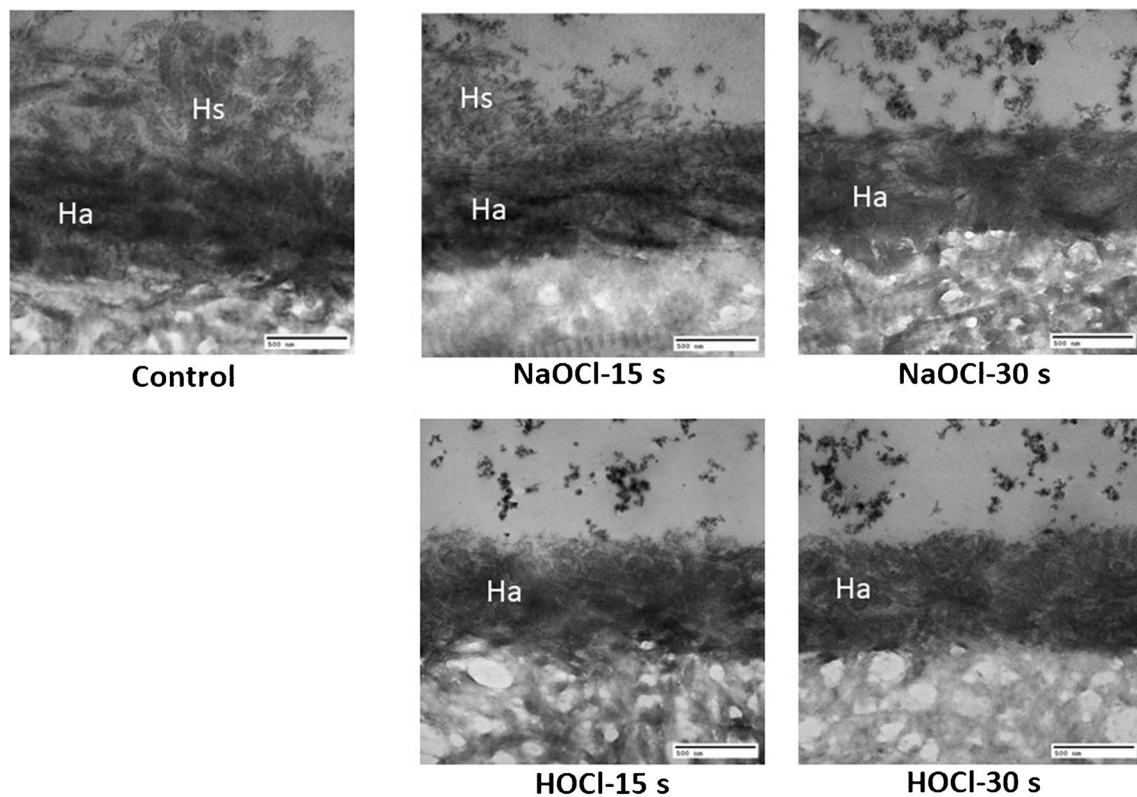
For the HOCl solution, Comfosy (Haccpper Advance), increasing the chlorine concentration decreases the pHs of the solutions (7.0, 6.7, and 6.4 of pH in 50, 100, and 200 ppm Comfosy, respectively) [19••]. Higher chlorine concentrations (100 and 200 ppm) in Comfosy, cause a slight reduction in treated dentin surface pH values, which might partially demineralize the dentin surface because these are slightly lower than the dentin critical pH (pHs 6.5–6.7) [19••]. On the other hand, 50 ppm Comfosy does not change the dentin surface pH, regardless of the application time (5, 15, or 30 s) [19••]. Fifteen-second and 30 s applications of 50 ppm HOCl solution (pH 7.0; Comfosy) on smear layer-covered dentin surface has a comparable deproteinizing ability to a similar duration treatment with 6% NaOCl solution, in which the mineral to matrix

ratio increased at the smear layer-covered dentin surface [8••]. The “top-down” removal of the organic phase from the mineralized dentin by NaOCl solution is diffusion-controlled and is both time and concentration-dependent [20], because the  $\text{OCl}^-$  ion is capable of infiltrating mineralized collagen [20]. Dentin exposed to 2% NaOCl for 4 h did not exhibit erosion but the presence of an intact dentin surface, and dentinal tubules of a normal dimension with barely visible lateral branches from the tubules were observed using TEM [20]. Following 15 and 30-s treatment with 6% NaOCl, appreciable morphological alterations could not be detected at the smear layer-covered dentin surface using SEM imaging [20]. These results would indicate that the deproteinization by the 15 and 30-s treatments of 6% NaOCl on smear layer-covered dentin surface does not extend to the underlying mineralized collagen. Presumably, deproteinization of the mineralized dentin by HOCl solution would be influenced by diffusion of HOCl molecules from the top surface of the smear layer-covered dentin, which is dependent upon the chlorine concentration and the application time. Therefore, 15 or 30-s deproteinization with 50 ppm HOCl solution would be more

limited within the smear layer than that occurring with a 6% NaOCl solution because of the lower concentration of chlorine and higher molecular weight of HOCl.

### Bonding Efficacy of Self-Etch Adhesive Systems to NaOCl-Deproteinized Dentin

Unfortunately, 15-s smear layer-deproteinization with 6% NaOCl slightly remained the hybridized smear layer at the adhesive interface of Clearfil SE Bond (Fig. 1) [8••] and could not improve the dentin bond strengths of the self-etch adhesives; Clearfil SE Bond (Kuraray Noritake Dental Inc.), Clearfil Protect Bond (Kuraray Noritake Dental Inc.), and Bond Force (Tokuyama Dental Corp) [19••, 21••]. On the other hand, it has been demonstrated that pretreatments with 6% NaOCl lasting more than 30s have an adverse effect on their dentin bond strengths [19••, 21••], and 30-s smear layer-deproteinization with 6% NaOCl severely increased nanoleakage formation at the adhesive interface of Clearfil SE Bond (Kuraray Noritake Dental Inc.) although the



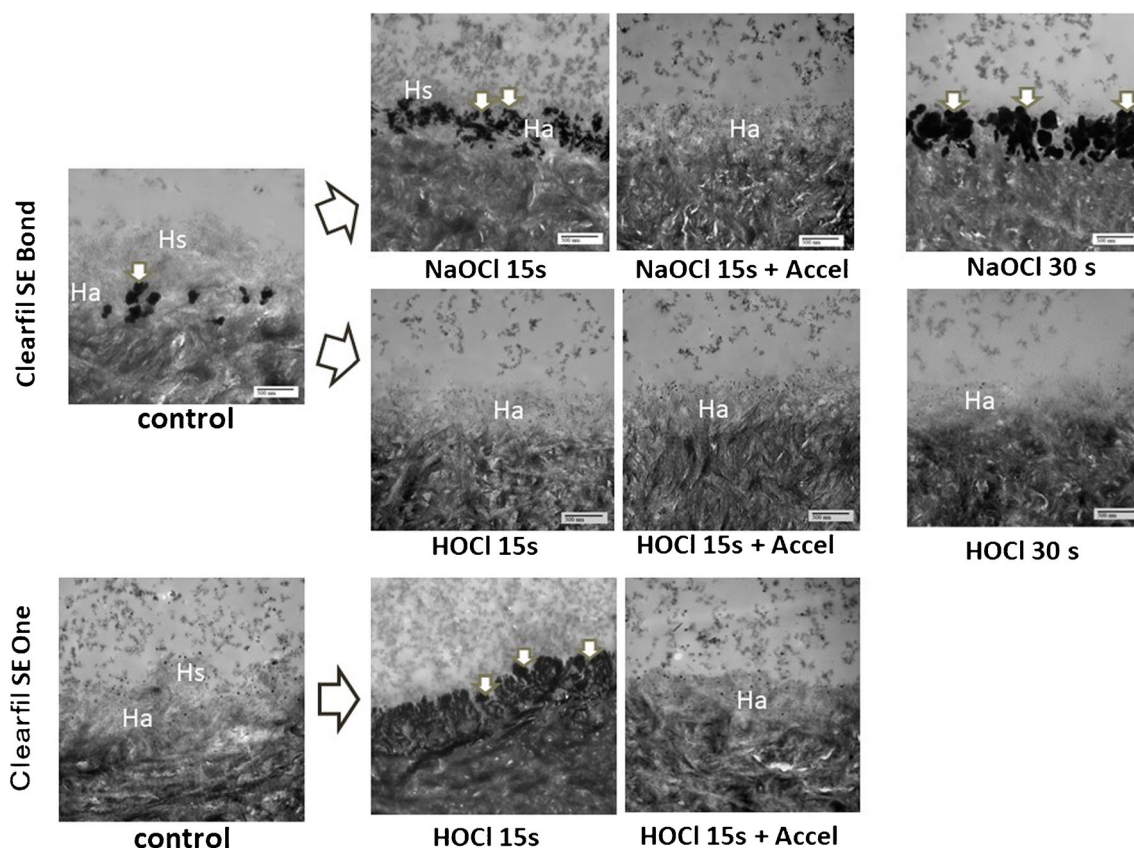
**Fig. 1** Demineralized TEM micrographs of the resin-dentin interface of Clearfil SE Bond with and without 6% NaOCl and 50 ppm HOCl deproteinizing. After pretreatment with 6% NaOCl and 50 ppm HOCl, bonded dentin slabs with Clearfil SE Bond were fixed and ultrathin-sectioned perpendicular to the adhesive interface. The specimens were demineralized and stained by 1% phosphotungstic acid followed by 2% uranyl acetate for examining the status of collagen at the resin-dentin

interface using a transmission electron microscope. The thick hybridized smear layer (Hs) can be seen on the authentic hybrid layer (Ha) in the no treatment group. The 15-s NaOCl deproteinizing slightly remained the hybridized smear layer, but the 30 s NaOCl deproteinizing could completely eliminate the hybridized smear layer. On the other hand, both 15 and 30-s HOCl deproteinizing could completely eliminate the hybridized smear layer. From Thanatvarakorn et al. [8••] with permission



hybridized smear layer was eliminated (Figs. 1 and 2) [8••]. This is thought to be because the reactive residual free-radicals generated by the oxidizing effect of NaOCl compete with the propagating vinyl free-radicals generated during light-activation of the adhesive, leading to incomplete polymerization by premature chain termination [22]. However, these compromised bond strengths to NaOCl-treated dentin can be recovered by applying 10% sodium ascorbate solution for longer than 60 s before the adhesive procedure [22], because it can interact with the by-products of NaOCl [23], resulting in neutralization and reversal of the oxidizing effect of the NaOCl-treated dentin surface [23]. In addition to this method, using a scrubbing technique for applying self-etch adhesives to NaOCl-treated dentin was reported to have a reversal effect on the compromised bonding because it could effectively remove the oxidized-byproducts on NaOCl-treated dentin as well as dentin smear layer [24].

Prasansuttiporn et al. evaluated the effect of three reducing agents/anti-oxidants: 10% sodium ascorbate, 100  $\mu$ M rosmarinic acid, and p-toluenesulfonic acid sodium salt (Accel; Sun Medical) solutions on the bond strengths of a self-etch adhesive (Clearfil Protect Bond, (Kuraray Noritake Dental Inc.)) to pretreated dentin with 6% NaOCl for 30 s [25••]. Accel and rosmarinic acid solutions improved bond strengths to NaOCl-treated dentin with a shorter application time (5 or 10 s) more than the sodium ascorbate solution. The p-toluenesulfonic acid sodium salt containing in Accel is an efficient accelerator for polymerization of resin composites, of which the reducing ability can recover the residual oxidizing effects at NaOCl-treated dentin by a redox reaction. The subsequent Accel application to NaOCl-treatment can improve nanoleakage formations and completely eliminate them at the adhesive interface of Clearfil SE Bond to 15-s NaOCl-treated dentin (Fig. 2) [8••]. Rosmarinic acid



**Fig. 2** Silver-stained TEM micrographs of resin-dentin interfaces of Clearfil SE Bond and Clearfil SE One. After pretreatment with 6% NaOCl or 50 ppm HOCl, Clearfil SE Bond or Clearfil SE One was applied to dentin surface. The bonded dentin slabs were immersed in ammoniacal silver nitrate solution for 24 h and then in photo-developing solution for 8 h under a fluorescent light. After fixed with osmium tetroxide and embedded in epoxy resin, the specimens were ultrathin-sectioned perpendicular to the adhesive interface. The nanoleakage expression of the adhesive interface was examined using a transmission electron microscope. Clearfil SE Bond; 15-s NaOCl

treatment increased nanoleakage expression with silver deposits (arrow). The following application of Accel could completely reduce nanoleakage expression in the 15-s NaOCl-treated dentin. On the other hand, 15-s HOCl (50 ppm) treatment decreased nanoleakage expression. Clearfil SE One; 15-s HOCl (50 ppm) treatment increased nanoleakage expression with silver deposits (arrow). The following application of Accel could completely decrease nanoleakage expression. Ha authentic hybrid layer, Hs Hybridized smear layer. From Thanatvarakom et al. [8••, 9••] with permission

extracted from rosemary has cross-linking and MMPs-inhibitor abilities as well as a high antioxidant capacity. Host-derived matrix metalloproteinases (MMPs) in human dentin can cause degeneration of exposed dentin collagen fibrils within the hybrid layer, leading to the loss of hybrid layer integrity and the reduction of resin-dentin bond strengths over time [26, 27]. The MMP-inhibitor ability of rosmarinic acid could improve the stability of resin-dentin bonds. Prasansuttiaporn et al. demonstrated that application of rosmarinic acid and Accel could improve the bonding durability in vitro of a self-etch adhesive (Clearfil SE Bond (Kuraray Noritake Dental Inc.)) to normal as well as 30-s smear layer-deproteinized dentin with 6% NaOCl [28].

### Bonding Efficacy of Self-Etch Adhesive Systems to HOCl-Deproteinized Dentin

Both 15 and 30-s smear layer-deproteinization with 50 ppm HOCl solution has been shown not to change the dentin bond strengths of Clearfil SE Bond (Kuraray Noritake Dental Inc.) [21••]. However, TEM observation of the resin-dentin interface revealed that both 15 and 30-s smear layer-deproteinization with 50 ppm HOCl solution resulted in reduced nanoleakage expression in the hybrid layer and elimination of the hybridized smear layer (Figs. 1 and 2) [8••]. On the other hand, 15-s smear layer-deproteinization with 6% NaOCl could not completely eliminate the hybridized smear layer and increased nanoleakage expression in the hybrid layer (Figs. 1 and 2) [8••]. These findings can be attributed to the fact that 50 ppm HOCl solution has a higher capacity to dissolve organic components and is easily washed away from the treated surface, leading to less residual oxidizing effects.

On the other hand, when a one-step self-etch adhesive, Clearfil SE One (Kuraray Noritake Dental Inc.) was used, nanoleakage expression increased when smear layer deproteinization with 50 ppm HOCl solution was undertaken for 15 s but reduced following the application of Accel (Sun Medical) (Fig. 2). These results can be attributed to the different polymerization systems in the adhesive systems as a result of different polymerization catalysts and accelerators. Thanatvarakorn et al. demonstrated that smear layer-deproteinization using 50 ppm HOCl solution for 15 s followed by the application of Accel could improve the quality of resin-dentin interface of the one-step self-etch adhesives BeautiBond Multi (Shofu Dental Corp.) and Bond Force (Tokuyama Dental Corp.) by eliminating the hybridized smear layer and preventing nanoleakage formation in the hybrid layer [9••]. In the case of Clearfil SE One (Kuraray Noritake Dental Inc.) and Scotchbond Universal (3M), which both contain the functional monomer, MDP, significant increases in dentin bond strengths were observed but there were no

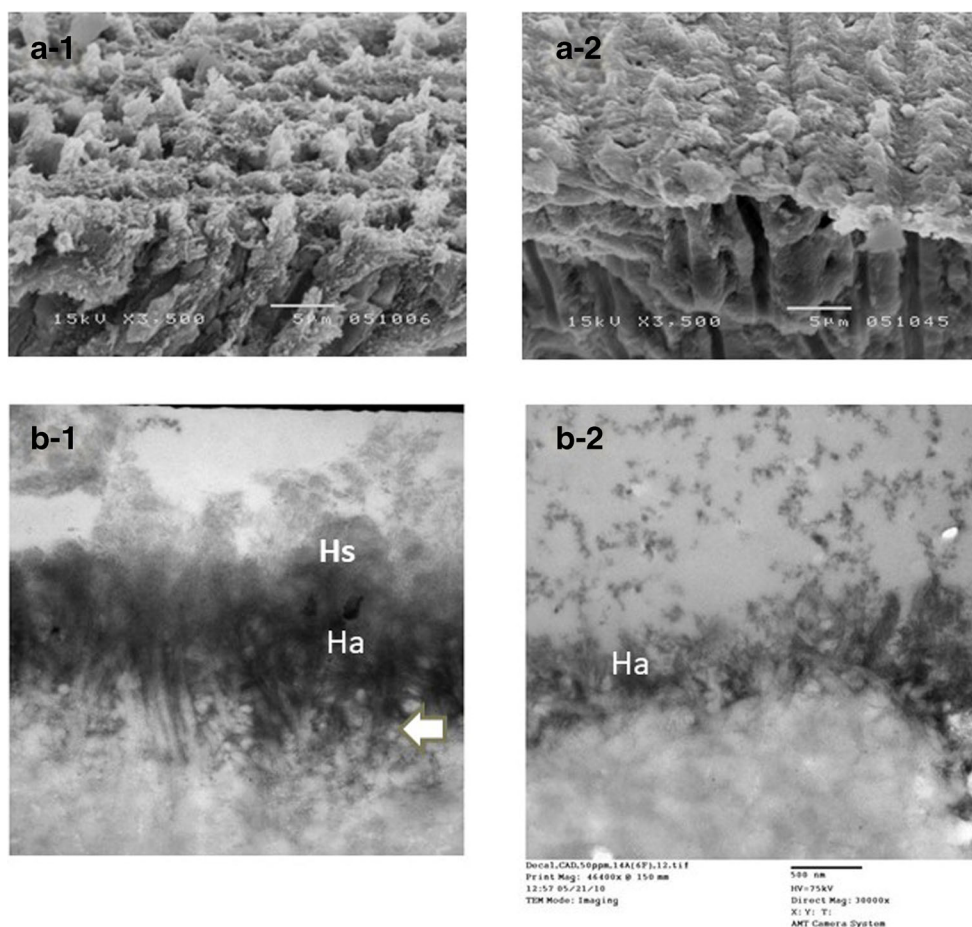
morphological improvement alterations at the adhesive interface [9••], which would be the result of enhanced chemical interactions to smear layer-deproteinized dentin through the formation of MDP-Ca salts [11]. Presumably, the positive effect of smear layer-deproteinization on the dentin bonding of self-etch adhesives would be influenced by the types of deproteinizing solutions and their application time, along with differences in the adhesive materials.

### Bonding to Caries-Affected Dentin

When investigating bond strengths of adhesive systems, the limited size and irregular shape of caries-affected dentin creates technical difficulties. The microtensile bond test allows to assess the bond strength to caries-affected dentin, because it permits tensile bond strengths to be measured in samples as small as 0.5 mm<sup>2</sup>. Using this method, it has been reported that bonding to caries-affected dentin results in lower bond strengths than normal dentin, regardless of the type of adhesive system (etch and rinse system or self-etch system; one-, two-, or three-step bonding procedure) [29••, 30–38], in which cohesive failure of specimens in dentin increases in resin-bonded caries-affected dentin [33, 34, 37]. A reduction in the cohesive strength of caries-affected dentin would be one of the reasons for lower bond strength values to caries-affected dentin compared with normal dentin [33]. Changes in the chemical and morphological characteristics of caries-affected dentin (i.e., alteration of apatite crystallites and intercrystalline spaces, reduction of mineral/organic ratio, increase in water content in intertubular dentin, presence of mineral deposits in dentinal tubules) would be also reasons for the inferior bonding performance to caries-affected dentin [39••]. In addition, caries-affected dentin produces thicker and more porous hybrid layers than normal dentin, because caries-affected dentin is partially demineralized and is more susceptible to acid etching, which results in the formation of a deeper demineralized zone (Fig. 3) [29••, 30–33, 37, 40].

### Improving the Adhesion of Self-Etch Adhesive Systems to Caries-Affected Dentin Using Smear Layer-Deproteinization with NaOCl and HOCl Solutions

The smear layer differs little in composition from the underlying dentin [41]. Therefore, the morphological and chemical structure of the smear layer formed on caries-affected dentin would be quite different from that formed from normal dentin, because caries-affected dentin is partially demineralized as a result of the cyclical process of demineralization and remineralization. Therefore, when compared with normal



**Fig. 3** **a** SEM micrographs of caries-affected dentin surface. After pretreatment with or without 50 ppm HOCl, morphological alterations of caries-affected dentin surface covered with smear layer were investigated using a scanning electron microscope. **a-1** No treatment group: the smear layer of caries-affected dentin was thick, irregular, and organic-rich with fibril-like structures. **a-2** 5-s HOCl (50 ppm) deproteinizing group: the smear layer was eroded and thinned due to dissolution of organic phase. From Kunawarote et al. [42] with permission. **b** Demineralized TEM micrographs of the adhesive interface of Clearfil SE Bond to caries-affected dentin. After

pretreatment with or without 50 ppm HOCl, Clearfil SE Bond to caries-affected dentin surface. The bonded specimens were demineralized and stained by 1% phosphotungstic acid followed by 2% uranyl acetate for examining the hybrid layer formation using a transmission electron microscope. **b-1** no treatment group: The thick hybridized smear layer (Hs) above the authentic hybrid layer (Ha) was seen and porous zone at the bottom of the authentic hybrid layer was observed (arrow). **b-2** 5-s HOCl (50 ppm) deproteinizing group: the thin and uniform authentic hybrid layer (Ha) was seen without the hybridized smear layer. From Nakajima et al. [39••] with permission

dentin, the smear layer on caries-affected dentin is thicker and appears to be enriched with organic components (Fig. 3) [20, 42]. The thicker gelatinized collagen layer within the smear layer of caries-affected dentin has been shown to hinder adhesive monomer infiltration and prevent a perfect seal at the resin-dentin interface, resulting in inferior adhesion for self-etch adhesive systems bonded to caries-affected dentin [39••].

Taniguchi et al. demonstrated that 15 s of 6% NaOCl smear layer-deproteinization could significantly improve the bond strengths of the two-step self-etch adhesive, Clearfil Protect Bond (Kuraray Noritake Dental Inc.) and the one-step self-etch adhesive, Bond Force (Tokuyama Dental Corp.), to caries-affected dentin, while it did not alter the bond strengths to normal dentin [20]. On the other hand, 30-s NaOCl smear layer-deproteinization did not affect the bond strengths to

caries-affected dentin, but reduced the bond strengths to normal dentin [20]. Deproteinization using NaOCl causes dramatic morphological changes on smear layer-covered caries-affected dentin, in which the smear layer is eroded and thinned, depending upon the application time [21••]. Removing the abundant organic phase within the smear layer of caries-affected dentin could offer an advantage for adhesive monomer infiltration. However, it has been speculated that applying NaOCl for 30 s or longer, results in a negative oxidizing effect within the treated dentin on the polymerization of the adhesive resin. A subsequent application of a reducing agent (Accel; Sun Medical) is effective in recovering this negative effect of the NaOCl-deproteinized dentin on polymerization, leading to increased bond strengths to caries-affected dentin as well as normal dentin [21••].



Kunawarote et al. described the effect of smear layer-deproteinization using 50 and 100 ppm HOCl solutions (Comfosy) on bonding to caries-affected dentin [42]. A five-second smear layer-deproteinization with 50 ppm HOCl solution significantly improved the bond strengths of Clearfil SE Bond (Kuraray Noritake Dental Inc.) to caries-affected dentin; however, 5-s smear layer-deproteinization with 6% NaOCl and 100 ppm HOCl solutions did not affect them [42]. TEM micrographs of the adhesive interface of caries-affected dentin revealed that the authentic hybrid layer had been thinned and the hybridized smear layer was absent (Fig. 3) [39••]. Therefore, 5-s smear layer-deproteinization with 50 ppm HOCl solution can improve the quality of the hybrid layer of caries-affected dentin using Clearfil SE Bond (Kuraray Noritake Dental Inc.) by eliminating the hybridized smear layer through the removal of disorganized/gelatinized collagen and enhancing adhesive monomer infiltration (Fig. 3) [39••], leading to the formation of a more stable adhesive interface at the caries-affected dentin surface. Smear layer-deproteinization in conjunction with self-etch adhesives is more effective in improving the bonding efficacy to caries-affected dentin than normal dentin because caries-affected dentin produces a thick organic-rich smear layer.

## Concluding Remarks

When caries is managed according to the concept of minimum intervention and the cavity is to be restored using an adhesive composite restoration, large areas of the cavity walls are composed of caries-affected dentin after removal of caries-infected dentin. However, caries-affected dentin produces inferior bonding performance and a poorer quality adhesive interface than normal dentin because of alteration of the morphological, chemical, and physical properties of caries-affected dentin [39••]. In addition, when the adhesive interface of caries-affected dentin is exposed to the oral environment, a poor quality hybrid layer would result in compromised dentin bonding durability due to hydrolysis at adhesive interface [40]. Caries-affected dentin still presents challenges as bonding substrate compared with normal dentin. However, there are only a few published studies on adhesion using caries-affected dentin as a bonding substrate because it is difficult to obtain many extracted human teeth with similar-sized carious dentin lesions. Therefore, bonding studies using artificial dentin caries induced by various methods have been conducted [43–48]; however, in some of these studies, preparation of smear layers on artificial caries-affected dentin surfaces has not been clarified. For self-etch adhesive systems, the characteristics of the smear layer is one of the important influencing factors on their dentin bonding performance and therefore smear layers should be prepared on the

chosen substrates when investigating the bonding performance of self-etch adhesive systems.

Unfortunately, many clinical studies have reported the failure cases of resin composite restoration, of which a main reason is secondary caries [49–52]. Considering secondary caries, wall lesion is mainly developed by interfacial gap formation between the resin composite and cavity walls, because saliva, fluid, and acids will enter the gap, and the cariogenic bacteria in the saliva will grow when the environment of gap is appropriate [53]. Thus, maintaining the integrity of adhesive interface in composite restoration would be an important factor for survival of the composite restored-tooth. However, even when an initial bonding with self-etch adhesives is sufficiently obtained, the adhesive interface would degrade over time [3] due to the nanoleakage formation at the interface and/or presence of hybridized smear layer with weak physical and chemical properties [8••, 9••], leading to deterioration of the sealing between resin composite and cavity walls. Smear layer-deproteinization with NaOCl and HOCl solutions can improve the quality of resin-dentin interface of self-etch adhesives through elimination of the hybridized smear layer, development of monomer infiltration, and enhancement of the chemical interaction between functional monomers and the underlying dentin [8••, 9••, 11, 12, 19••, 21••]. In the oral environment, the prepared dentin surface is occasionally contaminated by blood and saliva, and the dentin smear layer may trap bacteria. HOCl solution could be used as a cavity cleanser before placement of the restoration to eliminate these effects, which may impair the integrity of restored tooth, because it has a rapid and broad-spectrum antimicrobial activity against microorganisms with less irritating and sensitizing properties compared with NaOCl solution [13]. Smear layer-deproteinization with HOCl solution along with the subsequent application of an antioxidant/reducing agent could improve dentin bonding durability of self-etch adhesive systems [25••, 28], leading to enhancement of the longevity of composite restorations and protection against secondary caries and tooth fracture. In particular, with regard to bonding to caries-affected dentin with a thicker organic-rich smear layer, this potential improvement should be considered as a novel method for dentin surface modification in conjunction with self-etch adhesive materials [39••, 42].

## Compliance with Ethical Standards

**Conflict of Interest** The authors declare that they have no conflict of interest.

**Human and Animal Rights and Informed Consent** This article does not contain any studies with human or animal subjects performed by any of the authors.

## References

Papers of particular interest, published recently, have been highlighted as:

•• Of major importance

- Ogata M, Harada N, Yamaguchi S, Nakajima M, Pereira PNR, Tagami J. Effects of different burs on dentin bond strengths of self-etching priming systems. *Oper Dent*. 2001;26:370–7.
- Mahdan MHA, Nakajima M, Foxton RM, Tagami J. Combined effect of smear layer characteristics with hydrostatic pulpal pressure on dentine bond strength of HEMA-free and -containing one-step adhesives. *J Dent*. 2013;41:861–71.
- Shinoda Y, Nakajima M, Hosaka K, Otsuki M, Foxton RM, Tagami J. Effect of smear layer characteristics on dentin bonding durability of HEMA-free and HEMA-containing one-step self-etch adhesives. *Dent Mater J*. 2011;30(4):501–10.
- Tay FR, Pashley DH. Aggressiveness of contemporary self-etching systems. I: depth of penetration beyond dentin smear layers. *Dent Mater*. 2001;17:293–308.
- Suyama Y, Lührs AK, De Munck J, Mine A, Poitevin A, Yamada T, et al. Potential smear layer interference with bonding of self-etching adhesives to dentin. *J Adhes Dent*. 2013;15:317–24.
- Mine A, De Munck J, Cardoso MV, Van Landuyt KL, Poitevin A, Van Ende A, et al. Dentin-smear remains at self-etch adhesive interface. *Dent Mater*. 2014;30(10):1147–53.
- Spencer P, Wang Y, Walker MP, Swafford JR. Molecular structure of acid-etched dentin smear layers—in situ study. *J Dent Res*. 2001;80:1802–7.
- Thanatvarakorn O, Nakajima M, Prasansuttiporn T, Ichinose S, Foxton RM, Tagami J. Effect of smear layer deproteinizing on rein-dentine interface with self-etch adhesive. *J Dent*. 2014;42:298–304. **This article demonstrate that smear layer-deproteinizing with NaOCl and HOCl solution improves quality of the resin-dentin interface of a 2-step self-etch adhesive, eliminating hybridized smear layer, in which HOCl treatment was more effective than NaOCl treatment.**
- Thanatvarakorn O, Prasansuttiporn T, Thitthaweerat S, Foxton RM, Ichinose S, Tagami J, et al. Smear layer deproteinizing improves on bonding of one-step self-etch adhesives to dentin. *Dent Mater*. 2018;34(3):434–41. **This article clearly demonstrate that HOCl-smear layer-deproteinizing along with the subsequent application of a reducing agent improves dentin bonding of current 1-step self-etch adhesives.**
- Mountouris G, Silikas N, Eliades G. Effect of sodium hypochlorite treatment on the molecular composition and morphology of human coronal dentin. *J Adhes Dent*. 2004;6(3):175–82.
- Zhou L, Wang Y, Yang HY, Guo J, Tay RF, Huang C. Effect chemical interaction on the bonding strengths of self-etching adhesives to deproteinised dentine. *J Dent*. 2015;43:973–80.
- Kambara K, Nakajima M, Hosaka K, Takahashi M, Thanatvarakorn O, Ichinose S, et al. Effect of smear layer treatment on dentin bond of self-adhesive cements. *Dent Mater J*. 2012;31(6):980–7.
- Wang L, Bassiri M, Najafi R, Najafi K, Yang J, Khosrovi B, et al. Hypochlorous acid as a potential wound care agent: part I. Stabilized hypochlorous acid: a component of the inorganic armamentarium of innate immunity. *J Burns Wounds*. 2007;6:e5.
- Fukuzaki S. Mechanisms of actions of sodium hypochlorite in cleaning and disinfection processes. *Biocontrol Sci*. 2006;11(4):147–57.
- Albrich JM, McCarthy CA, Hurst JK. Biological reactivity of hypochlorous acid: implications for microbicidal mechanisms of leukocyte myeloperoxidase. *Proc Natl Acad Sci U S A*. 1981;78(1):210–4.
- White C. Handbook of chlorination and alternative disinfectants. New York: John Wiley & Sons, Inc.; 1999.
- Abdul-Baki AA. Pitfalls in using sodium hypochlorite as a seed disinfectant in C incorporation studies. *Plant Physiol*. 1974;53(5):768–71.
- Guentzel JL, Liang Lam K, Callan MA, Emmons SA, Dunham VL. Reduction of bacteria on spinach, lettuce, and surfaces in food service areas using neutral electrolyzed oxidizing water. *Food Microbiol*. 2008;25(1):36–41.
- Kunawarote S, Nakajima M, Shida K, Kitasako Y, Foxton RM, Tagami J. Effect of dentin pretreatment with mild acidic HOCl solution on microtensile bond strength and surface pH. *J Dent*. 2010;38(3):261–8. **This article represents the first study to highlight HOCl solution as a pretreatment agent with dentin bonding of self-etch adhesives.**
- Gu LS, Huang XQ, Griffin B, Bergeron BR, Pashley DH, Niu LN, et al. Primum non nocere—the effects of sodium hypochlorite on dentin as used in endodontics. *Acta Biomater*. 2017;61:144–56.
- Taniguchi G, Nakajima M, Hosaka K, Iwamoto N, Ikeda M, Foxton RM, et al. Improving the effect of NaOCl pretreatment on bonding to caries-affected dentin using self-etch adhesives. *J Dent*. 2009;37(10):769–75. **This article brings the first insight concerning smear layer-deproteinizing for bonding of self-etch adhesives to caries-affected dentin.**
- Lai SC, Mak YF, Cheung GS, Osorio R, Toledano M, Carvalho RM, et al. Reversal of compromised bonding to oxidized etched dentin. *J Dent Res*. 2001;80:1919–24.
- Morris MD, Lee KW, Agee KA, Bouillaguet S, Pashley DH. Effects of sodium hypochlorite and RC-prep on bond strengths of resin cement to endodontic surfaces. *J Endod*. 2001;27:753–7.
- Prasansuttiporn T, Nakajima M, Foxton RM, Tagami J. Scrubbing effect of self-etching adhesives on bond strength to NaOCl-treated dentin. *J Adhes Dent*. 2012;14(2):121–7.
- Prasansuttiporn T, Nakajima M, Kunawarote S, Foxton RM, Tagami J. Effect of reducing agents on bond strength to NaOCl-treated dentin. *Dent Mater*. 2011;27(3):229–34. **This article provides important information related to reversal effect of antioxidant/reducing agents on compromised bonding to NaOCl-treated dentin.**
- Pashley DH, Tay F, Yiu C, Hashimoto M, Breschi L, Carvalho R. Collagen degradation by host-derived enzymes during aging. *J Dent Res*. 2004;83:216–21.
- Carrilho MR, Carvalho RM, de Goes MF, di Hipolito V, Geraldini S, Tay FR, et al. Chlorhexidine preserves dentin bond in vitro. *J Dent Res*. 2007;86:90–4.
- Prasansuttiporn T, Thanatvarakorn O, Tagami J, Foxton RM, Nakajima M. Bonding durability of a self-etch adhesive to normal vs smear layer-deproteinized dentin: effect of reducing agent and plant extract antioxidant. *J Adhes Dent*. 2017;19:253–8.
- Nakajima M, Sano H, Burrow MF, Tagami J, Yoshiyama M, Ebisu S, et al. Tensile bond strength and SEM evaluation of caries-affected dentin using dentin adhesives. *J Dent Res*. 1995;74:1679–88. **This article is a classical study about bonding to caries-affected dentin. It is the first study to reduce bond strength to caries-affected dentin with thick and poor hybrid layer formation.**
- Nakajima M, Ogata M, Okuda M, Tagami J, Sano H, Pashley DH. Bonding to caries-affected dentin using self-etching primers. *Am J Dent*. 1999;12:309–14.
- Nakajima M, Sano H, Zheng L, Tagami J, Pashley DH. Effect of moist vs. dry bonding to normal vs. caries-affected dentin using Scotchbond multi-purpose plus. *J Dent Res*. 1999;78:1298–303.
- Nakajima M, Sano H, Urabe I, Tagami J, Pashley DH. Bond strengths of single-bottle dentin adhesives to caries-affected dentin. *Oper Dent*. 2000;25:2–10.



33. Yoshiyama M, Tay FR, Doi J, Nishitani Y, Yamada T, Itou K, et al. Bonding of self-etch and total-etch adhesives to carious dentin. *J Dent Res*. 2001;81:556–60.
34. Ceballos L, Camejo DG, Fuentes MV, Osorio R, Toledano M, Carvalho RM, et al. Microtensile bond strength of total-etch and self-etching adhesives to caries-affected dentine. *J Dent*. 2003;31:469–77.
35. Arrais CAG, Giannini M, Nakajima M, Tagami J. Effect of additional and extended acid etching on bonding to caries-affected dentin. *Eur J Oral Sci*. 2004;112:458–64.
36. Nakajima M, Kitasako Y, Okuda M, Foxton RM, Tagami J. Elemental distributions and microtensile bond strength of adhesive interface to normal and caries-affected dentin. *J Biomed Mater Res Part B: Appl Biomater*. 2005;72B:268–75.
37. Say EC, Nakajima M, Senawongse P, Soyman M, Ozer F, Tagami J. Bonding to sound vs. caries-affected dentin using photo and dual-cure adhesive. *Oper Dent*. 2005;30:90–8.
38. Nakajima M, Hosaka K, Yamauti M, Foxton RM, Tagami J. Bonding durability of self-etching primer system to normal and caries-affected dentin under hydrostatic pulpal pressure in vitro. *Am J Dent*. 2006;19:147–50.
39. Nakajima M, Kunawarote S, Prasansuttiporn T, Tagami J. Bonding to caries-affected dentin. *Jpn Dent Sci Rev*. 2011;47(2):102–14. **This review article gives valuable and detailed information related to bonding mechanism to caries-affected dentin.**
40. Erhardt MCG, Toledano M, Osorio R, Pimenta LA. Histomorphologic characterization and bond strength evaluation of caries-affected dentin/resin interfaces: effect of long-term water exposure. *Dent Mater*. 2008;24:786–98.
41. Pashley DH, Ciucchi B, Sano H, Horner JA. Permeability of dentin to adhesive agents. *Quintessence Int*. 1993;24(9):618–31.
42. Kunawarote S, Nakajima M, Foxton RM, Tagami J. Pretreatment effect of mildly acidic hypochlorous acid on adhesion to caries-affected dentin using a self-etch adhesive. *Eur J Oral Sci*. 2011;119(1):86–92.
43. Joves GJ, Inoue G, Nakashima S, Sadr A, Nikaido T, Tagami J. Mineral density, morphology and bond strength of natural versus artificial caries-affected dentin. *Dent Mater J*. 2013;32:138–43.
44. De Carvalho FG, Puppim-Rontani J, Dos Santos RL, Carlo HL, Bonan PR, Garcia-Godoy F, et al. In vitro effect of *S. mutans* biofilm on fluoride/MDPB-containing adhesive system bonded to caries-affected primary dentin. *Am J Dent*. 2014;27(5):227–32.
45. Lenzi TL, Tedesco TK, Calvo AF, Ricci HA, Hebling J, Raggio DP. Does the method of caries induction influence the bond strength to dentin of primary teeth? *J Adhes Dent*. 2014;16(4):333–8.
46. Peixoto AC, Bicalho AA, Isolan CP, Maske TT, Moraes RR, Cenci MS, et al. Bonding of adhesive luting agents to caries-affected dentin induced by a microcosm biofilm model. *Oper Dent*. 2015;40(3):E102–11.
47. Nicoloso GF, Antoniazzi BF, Lenzi TL, Soares FZ, Rocha RO. Is there a best protocol to optimize bond strength of a universal adhesive to artificially induced caries-affected primary or permanent dentin? *J Adhes Dent*. 2016;18(5):441–6.
48. Follak AC, Miotti LL, Lenzi TL, Rocha RO, Soares F. Degradation of multimode adhesive system bond strength to artificial caries-affected dentin due to water storage. *Oper Dent*. 2018;43(2):E92–E101.
49. Collins CJ, Bryant RW, Hodge KL. A clinical evaluation of posterior composite resin restorations: 8-year findings. *J Dent*. 1998;26:311–7.
50. Opdam NJ, Bronkhorst EM, Roeters JM, Loomans BA. A retrospective clinical study on longevity of posterior composite and amalgam restorations. *Dent Mater*. 2007;23:2–8.
51. Kopperud SE, Tveit AB, Gaarden T, Sandvik L, Espelid I. Longevity of posterior dental restorations and reasons for failure. *Eur J Oral Sci*. 2012;120:539–48.
52. Gordan VV, Riley JL III, Geraldelli S, Rindal DB, Qvist V, Fellows JL, et al. Repair or replacement of defective restorations by dentists in the dental practice-based research network. *J Am Dent Assoc*. 2012;143:593–601.
53. Diercke K, Lussi A, Kersten T, Seemann R. Isolated development of inner (wall) caries like lesions in a bacterial-based in vitro model. *Clin Oral Investg*. 2009;13:439–44.