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Influence of caries infiltrant contamination on shear bond strength of different adhesives to dentin

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Keywords

Caries infiltrant; Dentin; Adhesive; Shear bond strength; Hydrochloric acid, Phosphoric acid

Abstract

Objectives: To analyze whether the contamination with a caries infiltrant system impairs the adhesive performance of etch&rinse and self-etching adhesives on dentin.

Materials and methods: Dentin contamination with the caries infiltrant system (Icon, DMG) was simulated by applying either hydrochloric acid (15% HCl, Icon Etch, 15 s), the resin infiltrant (Icon infiltrant, 4 min) or both prior to the application of the respective adhesives (each group n=10). In the control groups, the etch&rinse adhesive (Optibond FL, Kerr) and the self-etching adhesive (iBOND Self Etch, Hereaus) were applied without former contamination with the infiltrant system. Additionally, the adhesive performance of the resin infiltrant alone was tested. Shear bond strength of a nano-hybrid composite was analyzed after thermo-cycling (5000x, 5°-55°C) of the specimens and analyzed by ANOVA/Scheffé post-hoc tests ($p < 0.05$) and Weibull statistics. Failure mode was inspected under a stereomicroscope at 25× magnification.

Results: Contamination with the resin infiltrant alone did not impair shear bond strength, while contamination with hydrochloric acid or with hydrochloric acid and the resin infiltrant reduced shear bond strength (MPa) of the adhesives (Optibond FL: 20.5 ± 3.6 , iBOND Self Etch: 17.9 ± 2.6) significantly. Hydrochloric acid contamination increased the number of adhesive failures. The adhesive performance of the caries infiltrant system alone was insufficient.

Conclusion: The contamination with the caries infiltrant system impaired the shear bond strength of conventional dental adhesives.

Clinical relevance: Contamination of the caries infiltrant system on dentin should be avoided due to the detrimental effect of hydrochloric acid etching.

Introduction

Originally, the caries infiltration technique has been introduced to arrest non-cavitated caries lesions by sealing the diffusion pathways of demineralized enamel with a low-viscosity resin. Compared to dental adhesives or fissure sealants, TEGDMA-based infiltrants were optimized for rapid capillary penetration and are able to penetrate enamel caries lesions almost completely after removal of the intact surface layer by etching with hydrochloric acid.

However, in recent studies it was questioned whether the range of application of the caries infiltrant system could be also extended to cavitated lesions [1-3]. Paris et al. [3] showed that the caries infiltrant is able to penetrate most parts of demineralized enamel in lesions with cavitation, but not capable to fill up the cavitation itself. However, resin infiltration might be successfully combined with adhesive conditioning to allow for restoration of cavitated and infiltration of demineralized areas. Studies by Wiegand et al. [1] and Jia et al. [2] showed that the caries infiltrant system achieved the same bond strength on sound and demineralized enamel as conventional adhesives and did not impair but even enhanced the bond strength of the adhesives when applied in combination. These results indicate that cavitated initial enamel lesions could be successfully restored with composite, while in the same step demineralized enamel at the margin of the cavitation can be preserved by resin infiltration. However, as larger cavitated carious lesions usually also involve dentin, it is of clinical relevance to which extent the adhesion to dentin is affected by contamination with the caries infiltrant system, especially emphasizing that dentin adhesion is much more complex as compared to bonding to enamel due to its high organic content and high hydrophilicity.

The commercially available caries infiltration system comprises 15% hydrochloric acid for the removal of the outer enamel surface layer, ethanol for drying and a TEGDMA-

based resin for infiltration of the lesion. The aim of the present study was to analyze to which extent the bond strength of an etch&rinse and a self-etch adhesive on dentin is affected by contamination with the whole caries infiltrant system or single components (hydrochloric acid and TEGDMA-based resin). Moreover, the adhesive properties of the caries infiltrant system itself should be assessed.

The null hypothesis was that contamination by the infiltrant material or its components does not impair bonding to dentin and allows for comparable shear bond strengths as compared to the control treatments.

Material and methods

Specimen preparation

Cylindrical dentin specimens (6.6 mm in diameter, n = 110) were prepared from the roots of freshly extracted, non-damaged bovine incisors. The specimens were then embedded in chemically cured acrylic resin (ScandiQuick, ScanDia, Hagen, Germany) and ground flat with P400SiC paper (Buehler, Lake Bluff, USA). The cementum layer was completely removed and checked by stereomicroscope analysis (M3B, Wild, Heerbrugg, Switzerland).

Specimen allocation and bonding procedure

Specimens were randomly divided into 11 groups of ten specimens each. The compositions of the adhesives and the infiltrant resin system are listed in Table 1.

The dentin surface was intentionally contaminated with the caries infiltrant system (Icon, DMG, Hamburg, Germany) before the respective etch&rinse (Optibond FL, Kerr, California, USA) or self-etching adhesive (iBOND Self Etch, HeraeusKulzer, Hanau,

Germany) was applied. To analyze the potentially adverse effect of the different components of the infiltrant system separately and in combination, the contamination was simulated by applying either hydrochloric acid (15% HCl, Icon Etch), the resin infiltrant (Icon infiltrant) or both. Contamination with hydrochloric acid was restricted to 15 s to avoid severe over-etching of dentin [4].

In the control groups, the etch&rinse and self-etching adhesives were applied and light-cured (20 s, 800 W/cm², bluephase, IvoclarVivadent, Schaan, Liechtenstein) according to the manufacturers' recommendation without contamination with the caries infiltrant system. The simulated contamination and bonding procedures in the different groups are listed in Table 2.

Additionally, the adhesive properties of the infiltrant alone (without adhesive application) after etching with 15% hydrochloric (120 s or 15 s, respectively) or 37% phosphoric acid (15 s) were analyzed.

Composite application and shear bond measurements

A nano-hybrid composite (TetricEvoCeram, IvoclarVivadent, Schaan, Liechtenstein) was then applied to the dentin surface using a transparent plastic hollow cylinder with an inner diameter of 3 mm [1, 2, 5]. The composite was packed against the surface in a 2 mm thick increment, which was then light-cured for 60 s. Light intensity was confirmed by a radiometer (Optilux Model 100, SDS Kerr Danbury, USA) after each 10 specimens. Bonding procedures were carried out by one operator (LJ) throughout all experiments.

Prior to shear bond strength testing, specimens were submitted to thermocycling (Willytec, Gräfelfing, Germany, 5000 cycles, 5° to 55° C, dwell time: 20 s, transfer time: 10 s) [6, 7]. Shear bond strength was tested with an Universal Testing Machine (ZwickZ010, Zwick, Ulm, Germany). A shear force was applied to the adhesive interface through a chisel-shaped loading device at a crosshead speed of 1 mm/min parallel to the

dentin surface. Load at fracture was recorded and shear bond strength (σ) was calculated by a software (TestXpert 11.02, Zwick, Ulm, Germany) using the load at failure F (N) and the adhesive area A (mm^2): $\sigma = F/A$.

The debonded area was examined for failure mode analysis with a stereomicroscope at 25x magnification (M3B, Wild, Heerbrugg, Switzerland). Failure mode was considered as adhesive, if it occurred at the interface and as cohesive if at least parts of either dentin or composite were affected.

Statistical analysis

Mean shear bond strength (\pm standard deviation) for each group was computed. Statistical analysis of the contamination and the control groups was done by two-way ANOVA, factors being the adhesives and the kind of contamination. Within each adhesive, one-way ANOVA followed by Scheffé's post-hoc tests were performed to analyze differences between contamination groups ($p \leq 0.05$).

Furthermore, for the calculation of the Weibull statistics, the least square estimates of the modulus and characteristic bond strength were computed according to the mean rank plotting (SPSS Version 20, SPSS INC, Chicago, USA) [8]. This statistical program allows only the calculation of the absolute estimates but not of the 95% CI. Also, post-hoc tests for Weibull parameters could not be obtained, so that a statistical comparison between the tested groups was not possible.

Relative frequencies of cohesive failures in each group were calculated at 95% CI.

Shear bond strength data of the caries infiltrant system after etching with hydrochloric or phosphoric acid but without application of an adhesive were close to zero and were therefore excluded from the statistical analysis.

Results

The shear bond strengths of the control groups amounted to 20.5 ± 3.6 MPa (Optibond FL) and 17.9 ± 2.6 MPa (iBond Self Etch).

Two-way ANOVA showed that contamination mode, adhesive material and the interaction between both factors was significant with respect to shear bond strength. Within each adhesive, one-way ANOVA revealed significant differences between the contamination groups.

For both adhesives, contamination with hydrochloric acid significantly reduced the shear bond strength. Application of the resin infiltrant after etching with hydrochloric acid and prior to adhesive application slightly increased shear bond strength as compared to the hydrochloric acid etching alone, but values were still significantly lower than in the control groups. Contamination with the resin infiltrant alone did not hamper bonding strength of both adhesives (Figure 1).

The Weibull parameters are presented in Table 3. The characteristic strength values were highest for the control groups and Optibond FL applied on dentin contaminated with the resin infiltrant. Weibull modulus m ranged from 5.1 to 6.3 for the Optibond FL groups and from 2.5 to 8.8 for the iBOND Self Etch groups.

With regard to the failure mode, the frequency of cohesive failures was decreased by the contamination with hydrochloric acid and by the whole caries infiltrant system, but not by the resin infiltrant alone (Table 4)

The adhesive performance of the caries infiltrant alone (after etching with hydrochloric or phosphoric acid) was insufficient. Shear bond strength values amounted to 0.3 ± 0.7 MPa (15 s hydrochloric acid), 0.1 ± 0.3 MPa (120 s hydrochloric acid) and 0.5 ± 1.2 MPa (15 s phosphoric acid). All failures of these groups were adhesive.

Discussion

This in vitro study showed that the adhesive performance of an etch&rinse and a self-etching adhesive was significantly reduced by contamination with the caries infiltrant system (15% HCl etching followed or not by resin infiltrant application) but not by the TEGDMA-containing resin infiltrant alone. Our results highlighted that the hydrochloric acid etching rather than the contamination with the resin infiltrant is detrimental for the adhesive properties of the conventional adhesives. Therefore, the null hypothesis that contamination by the infiltrant material or its components does not impair bonding to dentin and allows for comparable shear bond strengths as compared to the control treatments is partly rejected. Moreover, the caries infiltrant alone did not exhibit any adhesive properties on dentin, while it generated similar shear bond strength compared to conventional adhesives on sound and demineralised enamel [1, 2].

Shear bond strength was tested on bovine dentin, which has been proposed as suitable alternative for human dentin although shear bond strength is slightly higher on bovine root dentin compared to human coronal dentin [9]. Moreover, it should be considered that shear bond strength values in the present study might be higher than in the presence of intrapulpal pressure simulation [10, 11]. However, as relative differences rather than absolute values are of interest, the use of bovine dentin in adhesion testing - even without simulation of intrapulpal pressure - is widely accepted [12-14].

While hydrochloric acid etching is essential for the removal of the surface layer of non-cavitated enamel lesions to allow for the penetration of the resin infiltrant into the body of the lesion [15], hydrochloric acid contamination of sound dentin was shown to induce a more severe demineralization even in concentrations below 1% [4] as compared to 37% phosphoric acid. As a consequence of over-etching, the thick layer of demineralised

collagen is not capable of being completely impregnated by adhesives containing monomers with relatively high viscosity, such as Bis-GMA [16, 17].

In the present study, the application time of hydrochloric acid was reduced from 2 min (as recommended by the manufacturer for removal of the enamel surface layer) to 15 s to simulate only a contamination and avoid significant over-etching, which can be considered as an adaptation of the etch&rinse protocol using phosphoric acid. Although the surface was not extensively dried in these groups to avoid collapse of the exposed dentin network, shear bond strength was significantly reduced for both adhesives, indicating an incomplete penetration of demineralised dentin. This assumption is also confirmed by the fact that solely adhesive failures were observed in these groups.

Contamination with the whole caries infiltrant system (hydrochloric acid etching, ethanol, resin infiltrant application) resulted in slightly improved bond strength of the adhesives compared to the hydrochloric acid contamination alone, probably due to the improved penetration depth of the resin infiltrant. Due to the low viscosity, high penetration capability [18] and relatively high application time (total 4 min), we assume that the TEGDMA-containing infiltrant penetrates the severely demineralized dentin to a higher extent compared to the conventional adhesives. Contamination with the whole caries infiltrant system also included ethanol application on the dentin surfaces. Moisture control by ethanol pretreatment might increase the shear bond strength of hydrophilic monomers, like TEGDMA, to dentin [19]. However, shear bond strength after contamination with the caries infiltrant system was still significantly reduced compared to the control groups.

Contamination with the resin infiltrant alone did not impair bonding strength of Optibond FL and iBOND Self Etch significantly. In case of Optibond FL, the resin infiltrant contamination was performed on phosphoric acid etched dentin, thus TEGDMA might infiltrate the collagen network to a higher extent than Optibond FL. Therefore, the

amount of cohesive failures was considerably increased compared to the dentin contaminated with hydrochloric acid. The pretreatment with the TEGDMA-containing infiltrant probably results in a oxygen-inhibited layer [20], which allows a chemical connection between the infiltrant and the adhesive. As a consequence, shear bond strength was slightly (although not significantly) increased as compared to the control.

In contrast to etch&rinse adhesives, self-etching adhesives do not require a separate etching step, thus the resin infiltrant could not infiltrate demineralised dentin but only cover the surface of untreated dentin. However, although the shear bond strength data suggest, that the application and polymerization of the resin infiltrant on the dentin surfaces did not impair the demineralizing and adhesive efficacy of the self-etching adhesive, the Weibull modulus m is remarkably low in this group indicating low reliability [21]. Also, the amount of cohesive failures is reduced compared to the control. Except for this group, the shear bond strength data were supported by the Weibull parameters showing highest characteristic strength values for the control groups and Optibond FL applied on dentin contaminated with the TEGDMA-based resin infiltrant, and similar Weibull moduli m between 5.1 and 8.8.

Generally, it has to be borne in mind that the whole surface of the dentin specimens and not only parts were contaminated, which is not necessarily occurring clinically. Due to the high wettability of TEGDMA-resins it is likely that the exposed dentin area of a cavity becomes completely contaminated when the infiltrant is applied, while the contamination with hydrochloric acid can be easier controlled due to the higher viscosity and green colour of the gel. Thus, clinically, the contamination with hydrochloric acid gel can be probably restricted to smaller areas, so that the dentin adhesion might be less affected and the overall effect on the adhesion of the restoration is limited.

In conclusion, contamination of dentin during conditioning of enamel margins of cavitated lesions should be avoided. The crucial factor affecting shear bond strength is hydrochloric acid etching of dentin but not resin infiltrant application.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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Table 1: Composition of the caries infiltrant and the adhesive systems

Product	Composition	Batch number	Manufacturer
Icon	Icon-Etch: hydrochloric acid, pyrogenic silicic acid, surface-active substances	635703	DMG, Hamburg, Germany
	Icon-Dry: 99% ethanol	633314	
	Icon-Infiltrant: TEGDMA-based resin matrix, initiators, additives	633139	
Optibond FL	Primer: HEMA, ethanol, GPDM, MMEP, water, CQ, BHT	3463213	Kerr, Orange, California, USA
	Adhesive: Bis-GMA, HEMA, GDMA, CQ, ODMAB, approximately 48wt% filled	3486699	
iBOND Self Etch	UDMA, 4-META, glutaraldehyde, acetone, water, photo-initiators, stabilizers	010104	Heraeus, Hanau, Germany
<p>Bis-GMA=bisphenol A diglycidyl methacrylate, TEGDMA=triethylene glycol dimethacrylate, HEMA=2-hydroxyl methacrylate, GPDM=glycerol phosphate dimethacrylate, MMEP=mono-2-methacryloyloxyethyl phthalate, CQ= camphorquinone, BHT=butylhydroxytoluene, GDMA=glycerol dimethacrylate, ODMAB=2-(ethylhexyl)-4-(dimethylamino)benzoate, UDMA=urethane dimethacrylate,4-META=4-mathacryloyloxyethyl trimellitate anhydride.</p>			

Table 2: Bonding procedures and simulated contamination in the different groups.

Group	Contamination	Etching	Infiltrant	Adhesive
Optibond FL	None (control)	37% H ₃ PO ₄ (15 s)*	-	Optibond FL primer (15 s), Optibond FL adhesive (15 s), light curing (20 s)
	HCl (Icon Etch)	15 % HCl (15 s)*	-	Optibond FL primer (15 s) Optibond FL adhesive (15 s), light curing (20 s)
	HCl (Icon Etch) and resin infiltrant (Icon infiltrant)	15 % HCl (15 s)	Ethanol (Icon Dry, 30 s),* Resin infiltrant (3 min), Light curing (40 s), Resin infiltrant (1 min), Light curing (40 s)	Optibond FL primer (15 s), Optibond FL adhesive (15 s), light curing (20 s)
	Resin infiltrant (Icon infiltrant)	37% H ₃ PO ₄ (15 s)*	Resin infiltrant (3 min), Light curing (40 s), Resin infiltrant (1 min), Light curing (40 s)	Optibond FL primer (15 s), Optibond FL adhesive (15 s), light curing (20 s)
iBOND Self Etch	None (control)	-	-	iBOND Self Etch (20s), light curing (20 s)
	HCl (Icon Etch)	15 % HCl (15 s)*	.	iBOND Self Etch (20s), light curing (20 s)
	HCl (Icon Etch) and resin infiltrant (Icon infiltrant)	15 % HCl (15 s)	Ethanol (Icon Dry, 30 s),* Resin infiltrant (3 min), Light curing (40 s), Resin infiltrant (1 min), Light curing (40 s)	iBOND Self Etch (20s), light curing (20 s)
	Resin infiltrant (Icon infiltrant)	-	Resin infiltrant (3 min), Light curing (40 s), Resin infiltrant (1 min), Light curing (40 s)	iBOND Self Etch (20s), light curing (20 s)

The components of the caries infiltrant system were applied with the smooth surface-tip provided by the manufacturer. The adhesives were applied with a microbrush. The resin infiltrant and the adhesives were applied with light brushing motion. Excess infiltrant was removed by 5 air-blowing prior to light-curing.

*The surface was gently air-dried for 5 s.

Table 3: Weibull parameters. Characteristic strength σ_0 (MPa) and Weibull modulus m in different groups

Adhesive	Contamination	Characteristic bond strength σ_0	Weibull modulus m
Optibond FL	none	22.0	6.3
	HCl	13.1	5.3
	HCl and resin infiltrant	16.6	6.0
	Resin infiltrant	26.7	5.1
iBOND Self Etch	none	19.0	7.6
	HCl	5.6	8.8
	HCl and resin infiltrant	13.7	6.7
	Resin infiltrant	16.5	2.5

Table 4: Adhesive and cohesive failures and relative frequency of cohesive failures (95% CI) in the different groups

Cohesive failures occurred only in dentin but not in the composite

Adhesive	Contamination	Number of failure		Relative frequency (%) of cohesive failures (95% CI)
		adhesive	cohesive	
Optibond FL	none	4	6	60 (26.2; 87.8)
	HCl	10	0	0 (0.0; 30.8)
	HCl and resin infiltrant	8	2	20 (2.5; 55.6)
	Resin infiltrant	0	10	100 (69.2; 100.0)
iBOND Self Etch	none	6	4	40 (12.2; 73.8)
	HCl	10	0	0 (0.0; 30.8)
	HCl and resin infiltrant	10	0	0 (0.0; 30.8)
	Resin infiltrant	8	2	20 (2.5; 55.6)

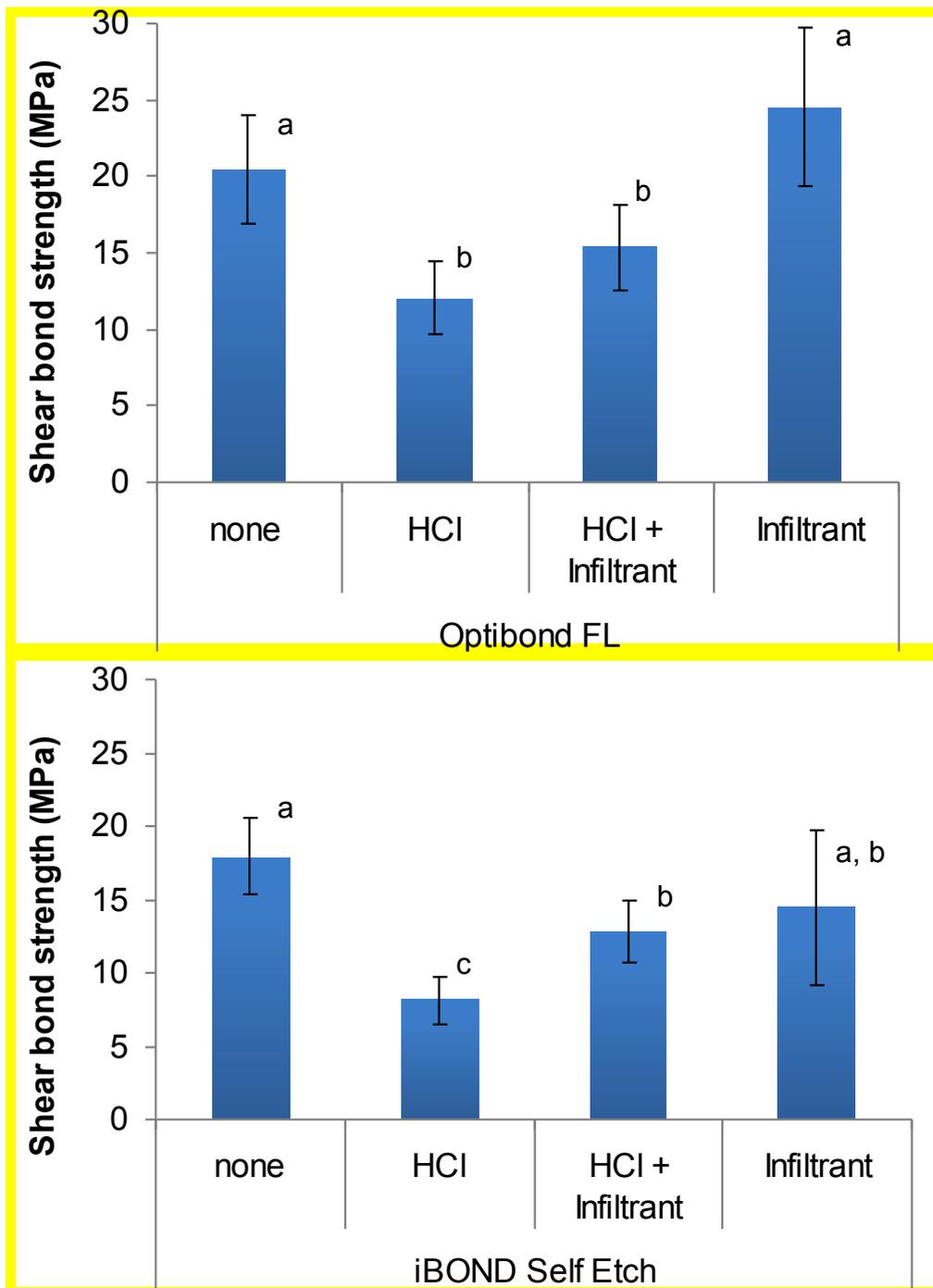


Figure 1

Shear bond strength (MPa, mean \pm standard deviation) of the different contamination groups in dentin treated with Optibond FL or iBOND Self Etch. Values which are not significantly different are marked with the same letter.