Acta Odontologica Scandinavica, 2011; 69: 182–192

informa healthcare

ORIGINAL ARTICLE

Effect of resin coating and occlusal loading on microleakage of class II computer-aided design/computer-aided manufacturing fabricated ceramic restorations: A confocal microscopic study

SHUZO KITAYAMA[1]*, NASSER A. NASSER[2]*, PETER PILECKI[2], RON F. WILSON[2], TORU NIKAIDO[1], JUNJI TAGAMI[1,3], TIMOTHY F. WATSON[2] & RICHARD M. FOXTON[2]

[1]*Cariology and Operative Dentistry, Department of Restorative Science, Graduate School, Tokyo Medical and Dental University, Tokyo, Japan,* [2]*Department of Restorative Dentistry, King's College London Dental Institute at Guy's, King's College and St. Thomas' Hospitals, King's College London, London, UK, and* [3]*Global Center of Excellence (GCOE) Program, International Research Center for Molecular Science in Tooth and Bone Diseases at Tokyo Medical and Dental University, Tokyo, Japan*

Abstract

Objective. To evaluate the effect of resin coating and occlusal loading on microleakage of class II computer-aided design/computer-aided manufacturing (CAD/CAM) ceramic restorations. ***Material and methods.*** Molars were prepared for an mesio-occlusal-distal (MOD) inlay and were divided into two groups: non-coated (controls); and resin-coated, in which the cavity was coated with a combination of a dentin bonding system (Clearfil Protect Bond) and a flowable resin composite (Clearfil Majesty Flow). Ceramic inlays were fabricated using the CAD/CAM technique (CEREC 3) and cemented with resin cement (Clearfil Esthetic Cement). After 24 h of water storage, the restored teeth in each group were divided into two subgroups: unloaded or loaded with an axial force of 80 N at a rate of 2.5 cycles/s for 250,000 cycles while stored in water. After immersion in 0.25% Rhodamine B solution, the teeth were sectioned bucco-lingually at the mesial and distal boxes. Tandem scanning confocal microscopy (TSM) was used for evaluation of microleakage. The locations of the measurements were assigned to the cavity walls and floor. ***Results.*** Loading did not have a significant effect on microleakage in either the resin-coated or non-coated group. Resin coating significantly reduced microleakage regardless of loading. The cavity floor exhibited greater microleakage compared to the cavity wall. TSM observation also revealed that microleakage at the enamel surface was minimal regardless of resin coating. In contrast, non-coated dentin showed extensive leakage, whereas resin-coated dentin showed decreased leakage. ***Conclusions.*** Resin coating with a combination of a dentin-bonding system and a flowable resin composite may be indicated prior to impression-taking when restoring teeth with CAD/CAM ceramic inlays in order to reduce microleakage at the tooth–resin interface.

Key Words: *Ceramic inlay, confocal microscopy, microleakage, occlusal loading, resin coating*

Introduction

All-ceramic restorations are characterized by enhanced esthetic properties, optimal integration to gingival tissues, and biocompatibility [1]. These restorations can be fabricated with a variety of systems, including the CEREC computer-aided design/computer-aided manufacturing (CAD/CAM) system (Sirona Dental Systems GmbH, Bensheim, Germany), which offers the dentist the opportunity to prepare, design, and fabricate a ceramic restoration at a single appointment, without the need to make impressions or provisional restorations or the need for dental laboratory support [2].

The adhesive-luting technique, including a dentin-bonding agent (DBA) and a resin cement, is now recommended for luting of many all-ceramic systems [3]. All-ceramic restorations cemented with adhesive materials have demonstrated superior fracture resistance and flexural strength compared with those cemented with conventional cements [3,4].

Correspondence: Shuzo Kitayama, Cariology and Operative Dentistry, Department of Restorative Science, Graduate School, Tokyo Medical and Dental University, 1-5-45 Yushima, Bunkyo-ku, Tokyo 113-8549, Japan. Tel: +81 3 5803 5483. Fax: +81 3 5803 0195. E-mail: nuevo_centro@yahoo.co.jp
*Both authors equally contributed to this work.

(Received 18 July 2010; accepted 16 November 2010)

ISSN 0001-6357 print/ISSN 1502-3850 online © 2011 Informa Healthcare
DOI: 10.3109/00016357.2010.549504

Although resin bonding between a tooth and a restoration is advocated for improving the retention, marginal adaptation, and inhibition of secondary caries [3,5], obtaining an effective and long-lasting marginal seal at the tooth-restorative material interface is still a great challenge. It has been reported that current resin cements do not always provide reliable bonding to dentin compared with dentin-bonding systems for direct resin–composite restorations [6,7]. A relatively weak bond of a resin cement may lead to poor adaptation and gap formation around the restoration [8], postoperative sensitivity [9], and reduced longevity of the restoration [6]. In addition, the clinical performance of the dentin bond is impaired by composite polymerization shrinkage and stresses due to thermal-induced dimensional changes [10]. Polymerization stress is increased when the cavity design shows a high configuration (C)-factor [11], as is the case with ceramic inlays. Therefore, the strength of the DBAs in resisting the polymerization shrinkage of resin cements on the dentin surface is of great importance for the prevention of microleakage.

In the early 1990s, a resin-coating technique was introduced for indirect restorations to minimize pulpal irritation and postoperative sensitivity [12]. Resin coating with a combination of a DBA and a low-viscosity resin composite (LVR) has been recommended for the prepared cavity immediately after tooth preparation, just before taking a final impression [13,14]. This technique also enables better bonding, sealing, and adaptation to dentin [13,14]. Therefore, adhesive luting following resin coating may be desirable to achieve long-term clinical success for ceramic restorations. Clinically, cemented restorations are subjected to repeated masticatory forces which may compromise the marginal integrity [15]; therefore, this cyclical loading should be replicated as closely as possible during testing of such restorations [16].

Leakage studies have been used as indicators of the marginal-sealing ability of restorations. However, to the authors' knowledge, no information is available regarding the effect of resin coating on the microleakage of class II CAD/CAM ceramic inlays fabricated with CEREC 3 under occlusal loading. Therefore, the purpose of this study was to evaluate the microleakage of class II CAD/CAM ceramic inlays using a resin-coating technique on the prepared cavity, as well as evaluating the influence of occlusal loading. The locations of the measured microleakage were assigned to the cavity walls and floor at the proximal boxes.

Material and methods

Specimens

Twenty-eight non-carious human lower third molars, extracted in accordance with local ethics approval rules (King's College London Dental Institute research ethics committee approval No. 04/Q0704/57) and with the informed consent of the patients, were used in the study. The teeth were stored in undisinfected tap water at 4°C, in order to avoid potential chemical media-induced artifacts [17], and used within 1 month of extraction. All selected third molars were of similar dimensions. The root of each specimen was dipped up to 1 mm below the cement–enamel junction in a two-part silicone model-duplicating material (SP8016 Hard; Bracon Dental Laboratory Products, Etchingham, UK) which was left to dry, so that each root was surrounded by a simulated periodontal membrane [18]. Then the root base of each tooth was embedded in acrylic resin (Cold Cure Modeling Acrylic; Mr Dental, Meadway, UK) for stabilization of the tooth.

Preparation design

Figure 1 summarizes schematically the design of the study. Each tooth was prepared to receive an MOD ceramic inlay using a regular flat-end taper diamond bur (ISO 169-714: L708.3M Two Striper FG Diam 13 mm; Abrasive Technology, Lewis Center, OH) under water coolant. The dimensions of the tapered preparations were 4 mm bucco-lingually, 3-mm deep at the isthmus, and 4-mm deep at the mesial and distal boxes and the boxes were 1.5 mm at the base towards the pulp. The cavities were prepared 1–1.5 mm above the cement–enamel junction (CEJ) and the boxes were prepared with butt margins gingivally (Figure 2). All the internal line angles were smoothed to reduce the possibility of stress concentrations.

All the prepared teeth were randomly divided into two groups of 14 teeth each, which were assigned to be resin-coated or not (Figure 1).

Materials

The resin-coating material and resin cement used in this study are listed in Table I. For the resin coating, a combination of a dentin-bonding system, Clearfil Protect Bond (Kuraray Medical, Tokyo, Japan), and a flowable composite, Clearfil Majesty Flow (Shade A3; Kuraray Medical), was used. Clearfil Protect Bond is a two-step, self-etch adhesive system composed of an antibacterial primer containing 12-methacryloyloxydodecylpyridinium bromide (MDPB) and a fluoride-releasing adhesive. A dual-cure resin cement, Clearfil Esthetic Cement (Kuraray Medical), was used for cementation. The self-etching primer, ED Primer II (Kuraray Medical), is the self-etching system used with Clearfil Esthetic Cement.

Resin coating

For the non-coated group, the prepared cavities were left as a control. For the resin-coated group, Clearfil

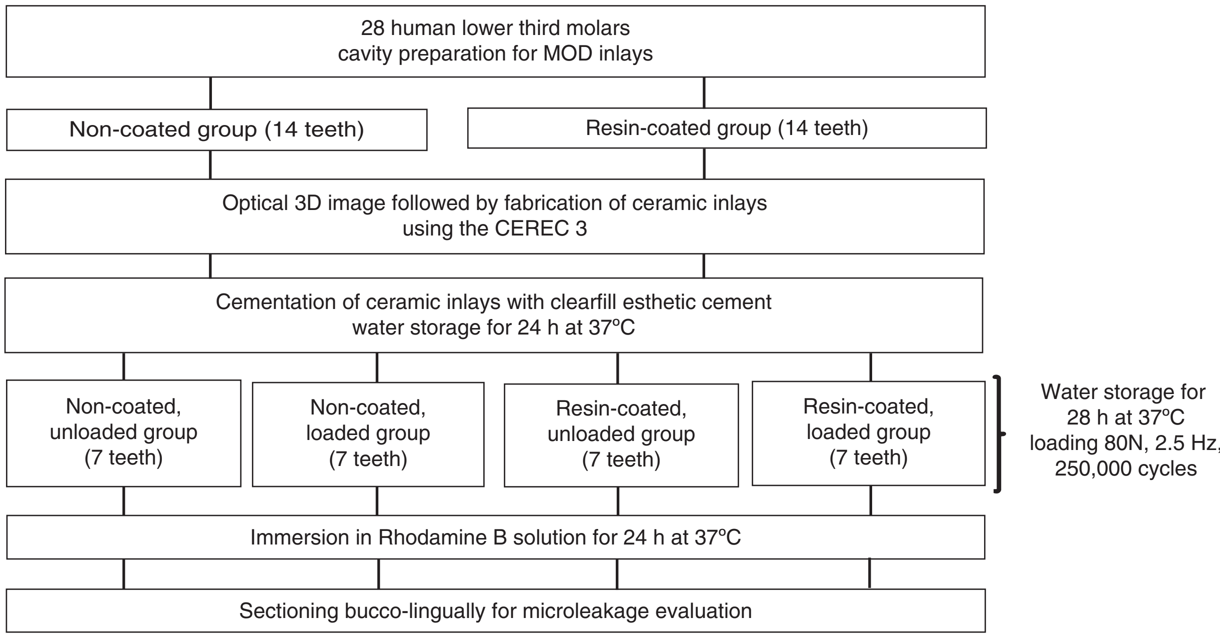

Figure 1. Schematic illustration of the design of the study.

Protect Bond was applied to the prepared cavities according to the manufacturer's instructions; the self-etching primer was applied to the dentin surface with a brush and left in place for 20 s. After drying the dentin surface with a mild air flow, the bonding agent was applied on the etched-primed dentin, gently air-flowed, and light-cured for 10 s. Thereafter, a thin layer of flowable composite, Clearfil Majesty Flow, was applied on the cured adhesive with a brush and light-cured for 20 s using a visible light-curing unit (Optilux 501; Sybron Kerr Corp., Orange, CA) with a curing intensity of 400 mW/cm^2. Contamination of the margins with the flowable resin was avoided (i.e. the margin was coated by the adhesive only) as sharp preparation margins are preferred for a mechanical milling procedure according to the manufacturer's instructions.

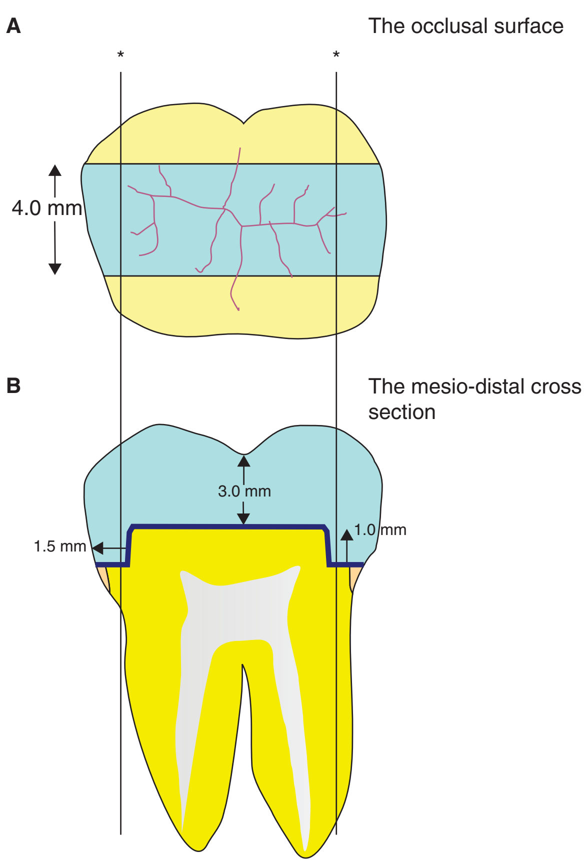

Figure 2. The occlusal surface (A) and the mesio-distal cross-section (B) of a tooth restored with a ceramic inlay. *Sectioning bucco-lingually at the proximal boxes.

Inlay fabrication with CAD/CAM

Imaging propellant (VITA CEREC Propellant; VITA Zahnfabric, Bad Säckingen, Germany) and powder (VITA CEREC Powder; VITA Zahnfabric) were applied to the tooth and optical 3D images were taken using the CEREC 3 milling unit. The CEREC 3 software was used to design 28 inlays, which were milled from machinable feldspathic ceramic blocks (CEREC-Blocs; Sirona Dental Systems GmbH) using the CEREC 3 milling unit.

Cementation procedures

The teeth were then cleaned with a cotton pellet soaked in isopropyl alcohol to remove the imaging powder. The fit of each ceramic inlay was checked and, when necessary, fitting adjustments were made by removing interferences from the inner surfaces of the inlays. The cementation procedures were performed according to the manufacturer's instructions.

Table I. Resin coating material and resin cement used in the study. All materials were manufactured by Kuraray Medical (Tokyo, Japan).

Material	Batch No.	Composition
Clearfil Protect Bond	00037A 00057A	Primer: MDP, MDPB, HEMA, hydrophilic dimethacrylate, water Bond: MDP, Bis-GMA, HEMA, hydrophobic dimethacrylate, dl-camphorquinone, *N,N*-diethanol-*p*-toluidine, silanated colloidal silica, surface-treated sodium fluoride
Clearfil Majesty Flow (Shade: A3)	00001A	Silanated barium glass filler, silanated colloidal silica, TEGDMA, hydrophobic aromaric dimethacrylate, dl-camphorquinone
K-Etchant Gel	00402A	Phosphoric acid, colloidal silica, pigments, water
Clearfil Ceramic Primer	00004E	3-Trimethoxysilylpropyl methacrylate, MDP, ethanol
Clearfil Esthetic Cement	00239A 00117A 0004BB 0004BB	ED Primer II A: HEMA, MDP, accelerator, water ED Primer II B: methacrylate monomers, initiator, accelerator, water Paste A: Bis-GMA, TEGDMA, other methacrylate monomers, silanated glass filler, colloidal silica Paste B: Bis-GMA, TEGDMA, other methacrylate monomers, silanated glass filler, silanated silica, colloidal silica, benzoyl peroxide, dl-camphorquinon, pigments

MDP = 10-methacryloyloxy-decyl dihydrogenphosphate; MDPB = 12-methacryloyloxydodecylpyridinium bromide; HEMA = 2-hydroxyethyl methacrylate; Bis-GMA = bisphenol A diglycidyl ether dimethacrylate; TEGDMA = triethyleneglycol dimethacrylate.

Regarding the pretreatment of the ceramic inlays, the internal surfaces were air-abraded with 50-µm alumina particles at 0.15 MPa for 10 s at a distance of 10 mm. Then, they were ultrasonically cleaned in distilled water for 5 min and air-dried. The internal surfaces were cleaned with 37% phosphoric acid (K-Etchant Gel; Kuraray Medical) for 10 s, rinsed, and air-dried. Following this, a silane-coupling agent (Clearfil Ceramic Primer; Kuraray Medical) was applied and air-dried.

Regarding the pretreatment of the tooth surface to be bonded, the non-coated and resin-coated teeth were treated in different ways. For the non-coated teeth, the prepared surfaces to be bonded were primed with ED Primer II for 30 s and air-dried. For the resin-coated teeth, the surfaces to be bonded were pre-treated according to the manufacturer's instructions: they were cleaned with 37% phosphoric acid (K-Etchant Gel) for 10 s, rinsed and air-dried in order to remove debris. Then, Clearfil Ceramic Primer was applied for silanization of the resin coating and air-dried. Following this, ED Primer II was applied for 30 s and air-dried.

Clearfil Esthetic Cement was placed onto the internal surfaces of inlays, they were then seated and excess cement was removed with a brush. The resin cement was light-cured for 60 s in total: 20 s each from occlusal, buccal, and lingual directions. The bonded specimens were stored in distilled water at 37°C for 24 h.

Occlusal loading

After the cementation procedure, teeth from both the non-coated and resin-coated groups were further divided into two subgroups of seven teeth each according to whether or not the tooth would be loaded (Figure 1). The teeth for loading were mounted in a customized loading machine driven by a linear actuator (LAL90; SMAC Europe Ltd., Horsham, UK). Oral mechanical stress was simulated at a rate of 2.5 cycles/s for 250,000 cycles with a load of 80 N in a water bath maintained at 37°C [19,20]. The load force was applied parallel to the long axis of the tooth at the central fissure using a 2-mm wide, round-ended, stainless-steel shaft [21]. The unloaded teeth were stored in distilled water at 37°C for an equivalent time span of ≈28 h.

Microleakage evaluation

The teeth were then carefully pulled out of the acrylic resin. For evaluation of microleakage, the teeth were sealed with two coats of nail varnish up to 1.0 mm below the cervical margins. They were then immersed in an aqueous 0.25% solution of Rhodamine B at 37°C for 24 h. After that, they were thoroughly rinsed in water and ultrasonically cleaned in distilled water for 5 min.

The teeth were sectioned twice bucco-lingually at the proximal boxes (mesial and distal) with a diamond blade (Extec Dia Wafer Blade; Extec Corp., Enfield, CT), providing two end sections per tooth (Figure 2), following polishing with 1000-grit silicon carbide paper (Struers Ltd, Solihull, UK) and ultrasonication in distilled water for 3 min. The two measurements obtained from one tooth were combined for microleakage evaluation.

A confocal microscope of the tandem scanning type (Noran Instruments, Middleton, WI) was used to focus below the surface smear layer created by sectioning and polishing. Samples were examined using a ×20/0.80 numerical aperture (NA) oil immersion objective in conjunction with a 4912 monochrome charge-coupled device (CCD) camera (Cohu Inc., San Diego, CA) and the image was relayed to a monitor. Dye penetration was observed by use of the appropriate fluorescence excitation and barrier filters (546/600 nm) and measured using a calibrated acetate sheet on the monitor screen. The locations of the measured microleakage were assigned to the cavity wall and floor (Figure 3). At each location, the microleakage at two interfaces was measured: the ceramic–cement interface; and the tooth–resin interface (i.e. the tooth–resin cement interface for the non-coated group and the tooth–resin coating interface for the resin-coated group). For all the observed interfaces, the length of the interface and the length of dye penetration were recorded, and the proportion of leakage was assessed for each location. High-resolution reflection and fluorescence images of the interfaces were taken using the same ×20 objective but in conjunction with an iXon 885 EM-CCD camera (Andor Technology, Belfast, UK).

Statistical analysis

The microleakage data were not normally distributed and could not be transformed. A non-parametric approach was taken to the description and analysis. Data were described using median and interquartile range and differences between materials and between loaded and unloaded conditions were analyzed separately for the cavity wall and floor using Mann–Whitney U-tests. Statistical significance for the above tests was predetermined at a significance level of 5%.

Results

The proportion of microleakage at the ceramic–resin and tooth–resin interfaces is shown in Tables II and III, respectively.

Microleakage at the ceramic–resin interface

Loading did not have a significant effect at each location in either the resin-coated or non-coated group ($P > 0.05$). Resin coating did not have a significant effect at each location in the loaded groups ($P > 0.05$), whereas it significantly decreased microleakage at each location in the unloaded groups ($P < 0.05$).

Microleakage at the tooth–resin interface

Again, loading did not have a significant effect at each location in either the resin-coated or non-coated group ($P > 0.05$). In contrast, resin coating significantly reduced microleakage at each location in both the loaded and unloaded groups ($P < 0.05$). The cavity floor exhibited greater microleakage compared to the cavity wall.

Tandem scanning microscopy images

Figures 4–6 show the tandem scanning microscopy (TSM) images of microleakage of the loaded specimens for each location. Minimum leakage was observed at the ceramic–resin interface in each

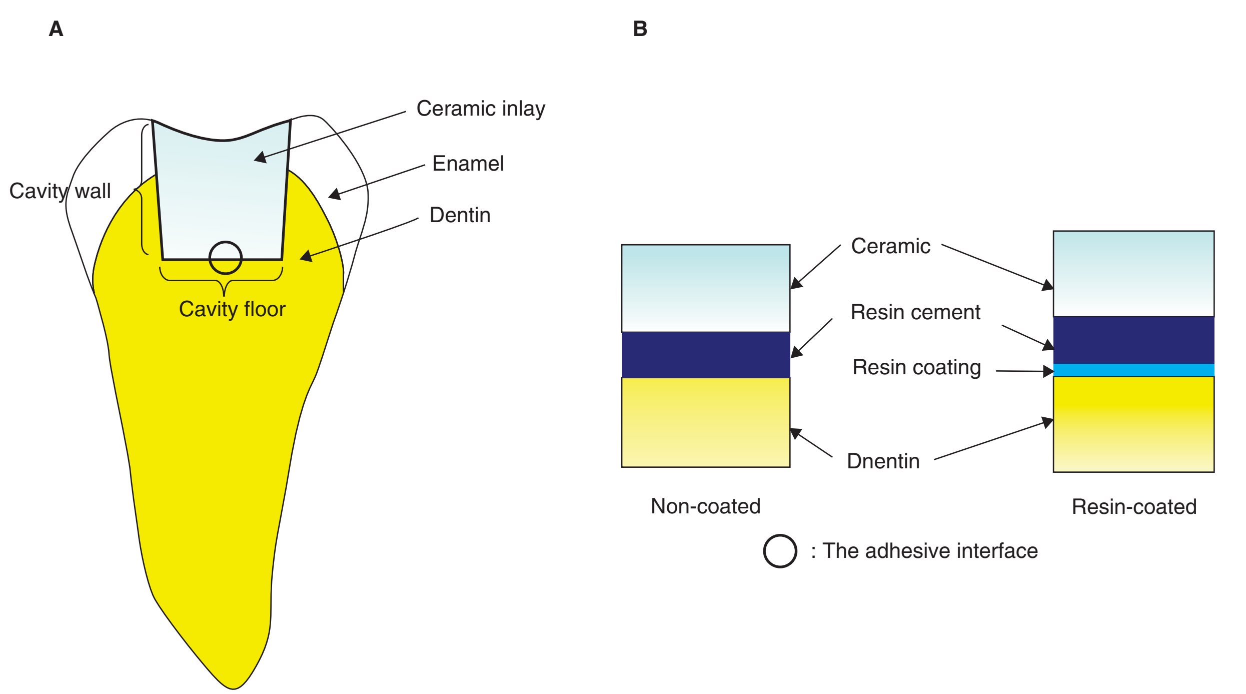

Figure 3. (A) The section for microleakage evaluation obtained by cutting through the proximal boxes. The circle indicates the adhesive interface. (B) The adhesive interfaces of non-coated and resin-coated specimens bucco-lingually.

Table II. Proportion of microleakage at the ceramic–resin interface (%). All values are medians, with inter-quartile ranges in parentheses.

	Cavity wall		Cavity floor	
	Unloaded	Loaded	Unloaded	Loaded
Non-coated	6.4[a] (2.6–8.9)	1.5 (0.7–8.3)	1.0[b] (0.3–28.5)	0 (0–2.9)
Resin-coated	0[a] (0–0)	0.7 (0–1.1)	0[b] (0–0)	0 (0–2.1)

[a,b]Values labeled with the same character are significantly different within columns ($P < 0.05$).

specimen (Figures 4 and 5). At the enamel surface in the cavity wall, the majority of the non-coated and resin-coated teeth showed minimal leakage (Figure 4). At the dentin surface in the cavity wall and floor, the majority of the non-coated teeth showed extensive leakage at the dentin–resin interface (Figures 5AB and 6AB). Many of the resin-coated teeth showed only a small amount of leakage (Figures 5CD and 6CD); however, some teeth showed extensive leakage (Figure 6EF). Fluorescent areas were identified within the adhesive layer of Clearfil Protect Bond in some images using the 546/600 nm filter set on the TSM (Figures 4D and 6F).

Discussion

This study was specifically designed to provide an accurate evaluation of the degree of microleakage occurring in the proximal walls of ceramic inlays bonded with a dual-cure resin cement. Of specific interest was the effect of resin coating applied to the prepared cavity before taking optical 3D images.

Previously, we examined the microleakage of CAD/CAM-fabricated all-ceramic crowns cemented using a resin-coating technique [21]. The prepared tooth was coated with a single-bottle adhesive (Clearfil Tri-S Bond; Kuraray Medical) followed by placement of a crown with resin cement, which failed to prevent microleakage due to the hydrophilicity of the adhesive [21]. Apparently, the effect on microleakage of using a more hydrophobic adhesive for the resin coating has yet to be examined. In the present study, a two-step self-etching primer–adhesive system, Clearfil Protect Bond, which is composed of a self-etching primer and an adhesive resin, was used as a DBA. The adhesive resin contains a hydrophobic methacrylate. It also contains 2-hydroxyethyl methacrylate (HEMA), which improves its permeability to dentin. However, the adhesive is solvent-free; in other words, it is not water-, acetone-, or ethanol-based. Therefore, the adhesive has more hydrophobic characteristics compared with an all-in-one adhesive [22]. Clearfil Protect Bond has shown good dentin bond durability and the potential for inhibiting secondary caries around restorations [23,24].

A single application of a DBA to the prepared cavity has been shown to protect the exposed dentin and prevent postoperative sensitivity [9,25]. Additionally, a previous study showed that additional application of an LVR on the cured adhesive significantly improved the sealing of the internal cavity margins [14] and the resin cement–dentin bond strength [13]. By coating the DBA with an LVR, the oxygen-inhibited, uncured layer of the DBA is eliminated [26]. Moreover, the uncured resin in the oxygen-inhibited layer will polymerize by light-curing the LVR, with the diffusion of free radicals from the LVR [27].

Ceramic inlays were air-abraded with alumina particles in order to increase the surface area for bonding [28], and cleaned by phosphoric acid followed by silanization, which is recommended by the manufacturer of the adhesive materials used. Hydrofluoric acid was shown to be an effective surface treatment of feldspathic ceramic for resin bonding [29]; however, it is a poisonous and caustic liquid that is extremely irritable to skin and lungs and is therefore unsuitable for use in a dental clinic. Since the present study simulated restoring the tooth at a single appointment, phosphoric acid was used instead.

A temporary filling was not applied to the prepared cavity as the ceramic inlay fabricated with the CEREC 3 milling unit can be cemented immediately after milling. Therefore, the possibility of contamination of the adhesive interface between the tooth (non-coated group) or resin-coating (resin-coated group) and resin cement by temporization was eliminated.

Table III. Proportion of microleakage at the tooth–resin interface (%). All values are medians, with inter-quartile ranges in parentheses.

	Cavity wall		Cavity floor	
	Unloaded	Loaded	Unloaded	Loaded
Non-coated	44.4[a] (41.4–49.9)	57.9[b] (48.0–60.2)	100[c] (100–100)	100[d] (100–100)
Resin-coated	8.8[a] (3.4–9.9)	12.5[b] (1.1–20.5)	39.7[c] (20.7–57.7)	53.1[d] (27.2–78.3)

[a–d]Values labeled with the same character are significantly different within columns ($P < 0.05$).

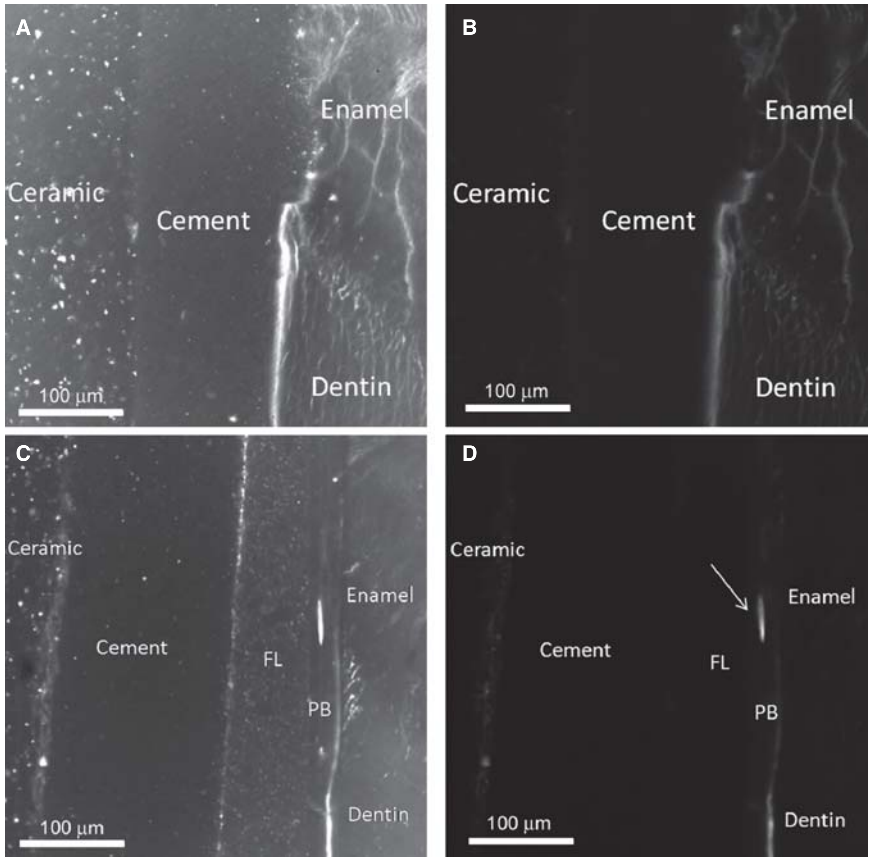

Figure 4. Confocal micrographs of microleakage of loaded specimens. Images of microleakage located at the enamel-dentin junction (EDJ) in the cavity wall. Panels (A) and (C) show reflection images of non-coated and resin-coated teeth, respectively. Fluorescence images show no leakage at the tooth–resin interface at the enamel surface, whereas leakage was observed at the dentin surface in both non-coated (B) and resin-coated teeth (D). Leakage at the ceramic–resin interface was minimal in both non-coated and resin-coated teeth. The arrow in panel (D) indicates a fluorescent area in PB, believed to result from the aqueous Rhodamine solution filling a space occupied by a sodium fluoride crystal. TSM; ×20/0.80 NA oil immersion objective. 546/– nm (A,C); 546/600 nm (B,D). Resin coating: PB+FL. PB = Clearfil Protect Bond; FL = Clearfil Majesty Flow.

The effect of mechanical loading was examined due to its potential for simulating mastication [15,30]. A force of 80 N was chosen as an average of the masticatory forces observed by Anderson [19]. The loading condition of 250,000 cycles has been verified as representing 1 year of clinical fatigue [20].

Confocal microscopy can be used to observe thin optical sections below the surface of a specimen [31]. Moreover, fluorophores can also act as markers for fluid penetration at the interface between the tooth and the restoration [31]. Microleakage studies performed with fluorescent dyes and examined using confocal microscopy may provide a more accurate description of restorative failure [32].

Loading did not significantly influence the microleakage at each interface in either the non-coated or resin-coated group. It has been reported that microleakage around restorations is accelerated by mechanical loading [15]. However, other studies have not found loading to have a significant effect on microleakage [16,30]. This disagreement may result from differences regarding the testing method, the materials used, and the magnitude and direction of load application. As in previous studies [15,30], the present study simulated only one of the complex movements involved in the *in-vivo* chewing process, and therefore does not simulate the reality of the clinical situation. These are the limitations of the current study and must be considered when interpreting the findings.

Microleakage at the ceramic–resin interface was <7% in each group. The levels of leakage reported for each group were sufficiently small as not to be of clinical significance, despite the statistical differences observed between some groups. This result is in agreement with the data of Schenke et al. [33], who investigated the marginal seal of partial ceramic crowns. The surface treatment of ceramic inlays applied in the present study has been shown to be effective when bonding feldspathic ceramic to resin [34]. In general, the ceramic–resin interface is indicated to show a greater integrity in the early bond seal than the tooth–resin interface.

Regarding the microleakage at the tooth–resin interface, resin coating significantly reduced microleakage at each location regardless of loading. Feilzer

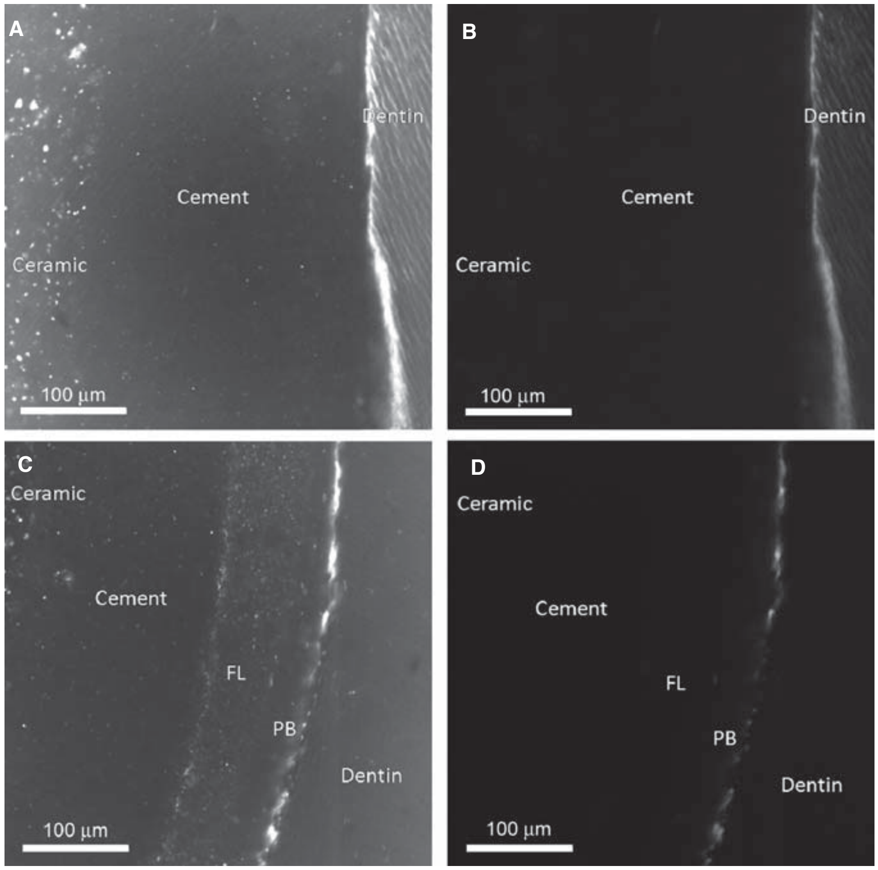

Figure 5. Confocal micrographs of microleakage at the dentin in the cavity wall (loaded specimens). Panels (A) and (C) show reflection images of non-coated and resin-coated teeth, respectively. Fluorescence image of a non-coated tooth (B) shows extensive leakage at the dentin–resin interface, whereas that of a resin-coated tooth (D) shows less leakage. Leakage at the ceramic–resin interface was not observed in either non-coated or resin-coated teeth. TSM; ×20/0.80 NA oil immersion objective. 546/– nm (A,C); 546/600 nm (B,D). Resin coating: PB+FL. PB = Clearfil Protect Bond; FL = Clearfil Majesty Flow.

et al. [35] have reported that the development of high polymerization stress in thin bonded layers is due to the unfavorable C factor in the conventional luting technique (non-coated group). The lining of the cavity with an LVR following preparation allows for free shrinkage of the thin layer due to the favorable ratio of the C factor, thus producing a primary stable bond to the tooth structure (resin-coated group) [33]. This bond is claimed to withstand polymerization shrinkage stresses that develop when the ceramic restoration is finally cemented to the resin-coated surface, with the resin coating acting as a stress absorber [14].

The microleakage was minimal at the enamel–resin interface. For the non-coated teeth, the prepared enamel was conditioned with ED Primer II followed by placement of Clearfil Esthetic Cement. For the resin-coated teeth, the prepared enamel was coated by the adhesive, Clearfil Protect Bond. The flowable composite was then applied to the cured adhesive except for the marginal area, in order to avoid altering the prepared margin. The results could indicate an ability to create an effectively bonded or sealed enamel–resin interface, i.e. enamel–Clearfil Esthetic Cement interface for non-coated teeth and enamel–Clearfil Protect Bond interface for resin-coated teeth. Additionally, it has been reported that bonding of the restorative materials to enamel is adequate to resist contraction stress [36].

For the non-coated group, the dentin was primed with ED Primer II, which is a one-step self-etching primer used in conjunction with Clearfil Esthetic Cement. It was reported that ED Primer II permits water-induced interfacial changes [37], which may explain extensive leakage at the dentin–resin interface. In contrast, for the resin-coated group, the prepared cavity was first coated with Clearfil Protect Bond, and water sorption of its predecessor, Clearfil SE Bond, has been reported to be significantly lower than that of one-step self-etch adhesives [38]. In addition, Chieffi et al. [22] demonstrated that the use of Panavia F with Clearfil Protect Bond produced a significantly higher dentin bond strength than with ED Primer II. Additionally, the DBA was coated by the flowable composite in the current study. The thickness of the flowable composite ranged between 50 and 170 µm, depending on the location. The smaller thickness of the flowable composite was found at

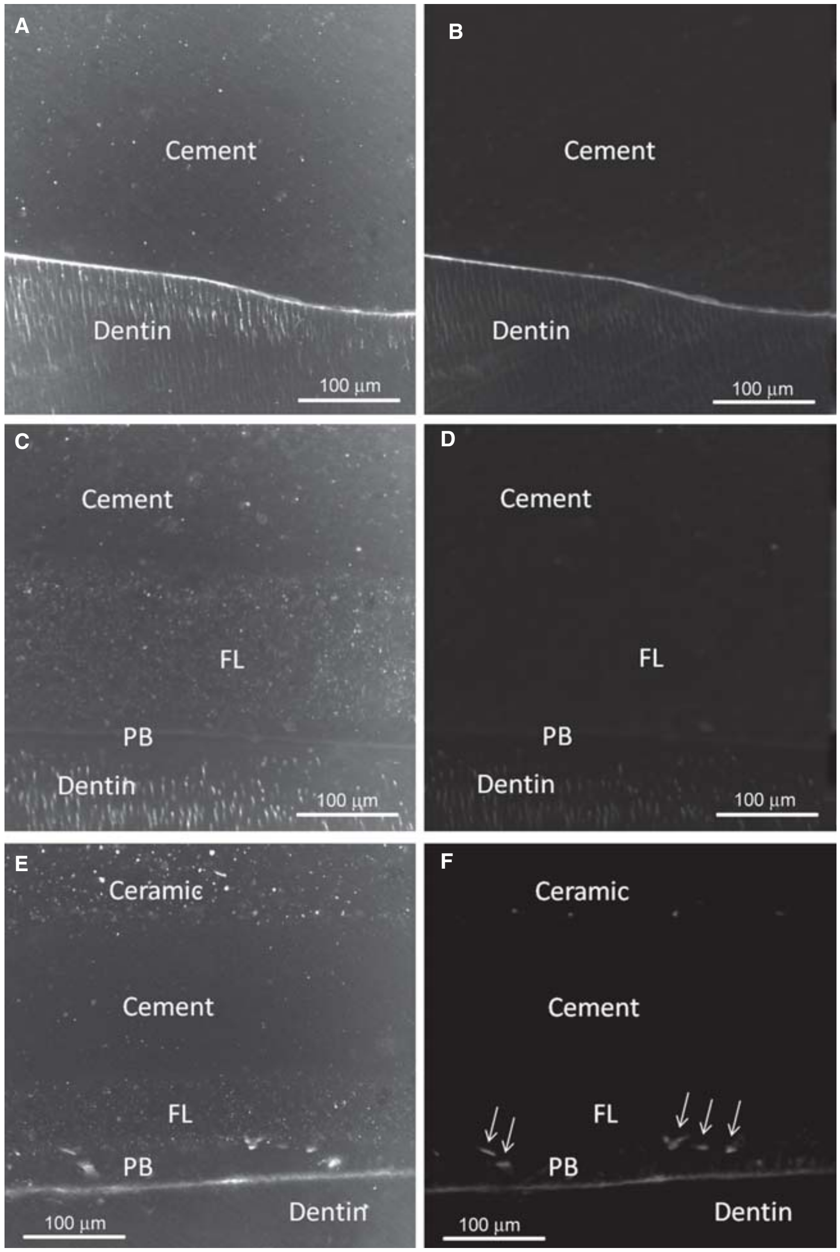

Figure 6. Confocal micrographs of microleakage at the cavity floor (loaded specimens). Image (A) shows a reflection image of a non-coated tooth, and images (C) and (E) show the same for resin-coated teeth. Fluorescence image of a non-coated tooth (B) shows extensive leakage at the dentin–resin interface. Fluorescence image of a resin-coated tooth (D) shows no leakage, whereas image (F) shows extensive leakage. The arrows in panel (F) indicate fluorescent areas in PB, believed to result from the Rhodamine solution filling spaces occupied by sodium fluoride crystals. TSM; ×20/0.80 NA oil immersion objective. 546/– nm (A,C); 546/600 nm (B,D). Resin coating: PB+FL. PB = Clearfil Protect Bond; FL = Clearfil Majesty Flow.

the cavity wall, whereas the thickness was higher at the cavity floor due to a gravitational effect. It is speculated that the dentin was well sealed by resin coating, thus reducing microleakage.

Since the thickness of enamel reduces as it approaches the cement-enamel junction (CEJ) [39], it could be postulated that the gingival margin lacked the integrity of the buccal, lingual, and occlusal margins. In addition, the thickness of the cement, which was observed using TSM, ranged between 40 and 350 μm for both the non-coated and resin-coated groups. Particularly, the deeper the cavity was, the thicker the cement was. Therefore, it is speculated that the amount of polymerization shrinkage was greater at the cavity floor than that at the cavity wall due to the difference in the volume of the cement. Furthermore, the distance of the light-curing unit to the cavity floor may have decreased the energy, reducing the degree of polymerization. Since better polymerization enhances adhesion [40], the lower conversion of monomer may have contributed to the poorer sealing obtained at the cavity floor compared to the cavity wall. These facts could explain why the cavity floor showed greater leakage than the cavity wall. Additionally, the differences in the location and orientation of dentinal tubules may have influenced the microleakage [41].

Donmez et al. [24] observed sodium fluoride (NaF) crystals in the adhesive layer of Clearfil Protect Bond. In the current study, similar features were found and, interestingly, they were fluorescent when viewed with the TSM using the 546/600 nm filter set. We postulate that NaF was released from the adhesive and then surrounded by the Rhodamine solution due to leakage. Since the dye is water-based, the NaF may have dissolved, resulting in the space it had occupied being filled by the dye. This phenomenon may be exclusive to Clearfil Protect Bond. Further study should be conducted to confirm this hypothesis.

It must be stressed that microleakage scored on sectioned restorations operates within the limitations of a 2D valuation of a 3D interface. Within these limitations, where discrete leakage in dentin is recorded at a similar depth to a complete line of dye in another specimen, it is arguable that the prognosis for both restorations will be the advent of secondary caries.

The importance of evaluating the influence of occlusal loading on bond quality is increasing, since the application of adhesive materials in posterior teeth has become very popular in daily practice. Therefore, it would be useful to conduct further studies utilizing different combinations of cycle repetition, load magnitude, and site of load application in order to identify relevant occlusal load simulations relating to clinical wear time points for these materials.

Within the limitations of this study, it can be concluded that resin coating with a combination of a dentin-bonding system and a flowable composite is effective in reducing microleakage at the tooth–resin interface of class II CAD/CAM ceramic inlays cemented with dual-cure resin cement, whether loaded or unloaded.

Acknowledgements

This study was supported in part by the Great Britain Sasakawa Foundation.

Declaration of interest: The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

References

[1] Fradeani M, Redemagni M. An 11-year clinical evaluation of leucite-reinforced glass-ceramic crowns: a retrospective study. Quintessence Int 2002;33:503–10.

[2] Zimmer S, Gohlich O, Ruttermann S, Lang H, Raab WH, Barthel CR. Long-term survival of Cerec restorations: a 10-year study. Oper Dent 2008;33:484–7.

[3] Burke FJ, Fleming GJ, Nathanson D, Marquis PM. Are adhesive technologies needed to support ceramics? An assessment of the current evidence. J Adhes Dent 2002; 4:7–22.

[4] Burke FJ. The effect of variations in bonding procedure on fracture resistance of dentin-bonded all-ceramic crowns. Quintessence Int 1995;26:293–300.

[5] Rosenstiel SF, Land MF, Crispin BJ. Dental luting agents: A review of the current literature. J Prosthet Dent 1998;80: 280–301.

[6] Burrow MF, Nikaido T, Satoh M, Tagami J. Early bonding of resin cements to dentin–effect of bonding environment. Oper Dent 1996;21:196–202.

[7] Okuda M, Nikaido T, Maruoka R, Foxton RM, Tagami J. Microtensile bond strengths to cavity floor dentin in indirect composite restorations using resin coating. J Esthet Restor Dent 2007;19:38–46.

[8] Piwowarczyk A, Lauer HC, Sorensen JA. Microleakage of various cementing agents for full cast crowns. Dent Mater 2005;21:445–53.

[9] Christensen GJ. Resin cements and postoperative sensitivity. J Am Dent Assoc 2000;131:1197–9.

[10] Van Meerbeek B, Perdigao J, Lambrechts P, Vanherle G. The clinical performance of adhesives. J Dent 1998;26:1–20.

[11] Feilzer AJ, de Gee AJ, Davidson CL. Setting stress in composite resin in relation to configuration of the restoration. J Dent Res 1987;66:1636–9.

[12] Inokoshi S, Shimada Y, Fujitani M, Otsuki M, Shono T, Onoe N, et al. Monkey pulpal response to adhesively luted indirect resin composite inlays. Oper Dent 1995;20: 111–18.

[13] Jayasooriya PR, Pereira PN, Nikaido T, Tagami J. Efficacy of a resin coating on bond strengths of resin cement to dentin. J Esthet Restor Dent 2003;15:105–13.

[14] Jayasooriya PR, Pereira PN, Nikaido T, Burrow MF, Tagami J. The effect of a "resin coating" on the interfacial adaptation of composite inlays. Oper Dent 2003;28:28–35.

[15] da cunha Mello FS, Feilzer AJ, de Gee AJ, Davidson CL. Sealing ability of eight resin bonding systems in a Class II restoration after mechanical fatiguing. Dent Mater 1997;13: 372–6.

[16] Gu XH, Kern M. Marginal discrepancies and leakage of all-ceramic crowns: influence of luting agents and aging conditions. Int J Prosthodont 2003;16:109–16.

[17] Titley KC, Chernecky R, Rossouw PE, Kulkarni GV. The effect of various storage methods and media on shear-bond strengths of dental composite resin to bovine dentine. Arch Oral Biol 1998;43:305–11.

[18] Hayashi M, Sugeta A, Takahashi Y, Imazato S, Ebisu S. Static and fatigue fracture resistances of pulpless teeth restored with post-cores. Dent Mater 2008;24:1178–86.

[19] Anderson D. Measurement of stress in mastication. II. J Dent Res 1956;35:671–3.

[20] Sakaguchi RL, Douglas WH, Delong R, Pintado MR. The wear of a posterior composite in an artificial mouth: a clinical correlation. Dent Mater 1986;2:235–40.

[21] Kitayama S, Pilecki P, Nasser AN, Bravis T, Wilson RF, Nikaido T, et al. Effect of resin coating on adhesion and microleakage of computer-aided design/computer-aided manufacturing fabricated all-ceramic crowns after occlusal loading: a laboratory study. Eur J Oral Sci 2009; 117:454–62.

[22] Chieffi N, Chersoni S, Papacchini F, Vano M, Goracci C, Davidson CL, et al. The effect of application sustained seating pressure on adhesive luting procedure. Dent Mater 2007;23: 159–64.

[23] Tsuchiya S, Nikaido T, Sonoda H, Foxton RM, Tagami J. Ultrastructure of the dentin-adhesive interface after acid-base challenge. J Adhes Dent 2004;6:183–90.

[24] Donmez N, Belli S, Pashley DH, Tay FR. Ultrastructural correlates of in vivo/in vitro bond degradation in self-etch adhesives. J Dent Res 2005;84:355–9.
[25] Momoi Y, Akimoto N, Kida K, Yip KH, Kohno A. Sealing ability of dentin coating using adhesive resin systems. Am J Dent 2003;16:105–11.
[26] Sanares AM, Itthagarun A, King NM, Tay FR, Pashley DH. Adverse surface interactions between one-bottle light-cured adhesives and chemical-cured composites. Dent Mater 2001; 17:542–56.
[27] Rueggeberg FA, Margeson DH. The effect of oxygen inhibition on an unfilled/filled composite system. J Dent Res 1990; 69:1652–8.
[28] Ayad MF, Fahmy NZ, Rosenstiel SF. Effect of surface treatment on roughness and bond strength of a heat-pressed ceramic. J Prosthet Dent 2008;99:123–30.
[29] El Zohairy AA, De Gee AJ, Hassan FM, Feilzer AJ. The effect of adhesives with various degrees of hydrophilicity on resin ceramic bond durability. Dent Mater 2004;20:778–87.
[30] Darbyshire PA, Messer LB, Douglas WH. Microleakage in class II composite restorations bonded to dentin using thermal and load cycling. J Dent Res 1988;67:585–7.
[31] Watson TF, Boyde A. Tandem scanning reflected light microscopy: applications in clinical dental research. Scanning Microsc 1987;1:1971–81.
[32] Watson TF, Boyde A. Confocal light microscopic techniques for examining dental operative procedures and dental materials. A status report for the American Journal of Dentistry. Am J Dent 1991;4:193–200.
[33] Schenke F, Hiller KA, Schmalz G, Federlin M. Marginal integrity of partial ceramic crowns within dentin with different luting techniques and materials. Oper Dent 2008;33:516–25.
[34] Kamada K, Yoshida K, Atsuta M. Effect of ceramic surface treatments on the bond of four resin luting agents to a ceramic material. J Prosthet Dent 1998;79:508–13.
[35] Feilzer AJ, de Gee AJ, Davidson CL. Increased wall-to-wall curing contraction in thin bonded resin layers. J Dent Res 1989;68:48–50.
[36] Hilton TJ. Can modern restorative procedures and materials reliably seal cavities? In vitro investigations. Part 2. Am J Dent 2002;15:279–89.
[37] Carvalho RM, Pegoraro TA, Tay FR, Pegoraro LF, Silva NR, Pashley DH. Adhesive permeability affects coupling of resin cements that utilise self-etching primers to dentine. J Dent 2004;32:55–65.
[38] Malacarne J, Carvalho RM, de Goes MF, Svizero N, Pashley DH, Tay FR, et al. Water sorption/solubility of dental adhesive resins. Dent Mater 2006;22:973–80.
[39] Dietschi D, Scampa U, Campanile G, Holz J. Marginal adaptation and seal of direct and indirect Class II composite resin restorations: an in vitro evaluation. Quintessence Int 1995;26:127–38.
[40] Yanagawa T, Finger WJ. Relationship between degree of polymerization of resin composite and bond strength to Gluma-treated dentin. Am J Dent 1994;7:157–60.
[41] Phrukkanon S, Burrow MF, Tyas MJ. The effect of dentine location and tubule orientation on the bond strengths between resin and dentine. J Dent 1999;27:265–74.