



## Effect of Rinsing Water Temperature on Microleakage of Dental Composite Restorations: An In-Vitro Study

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Article Info	ABSTRACT
<p><b>Article type:</b> Original Article</p> <hr/> <p><b>Article History:</b> Received: 05 Feb 2025 Accepted: 10 May 2025 Published: 01 Feb 2026</p> <hr/> <p><b>* Corresponding author:</b> Department of Operative Dentistry, Faculty of Dentistry, Rafsanjan University of Medical Sciences, Rafsanjan, Iran  Email: <a href="mailto:Niloufarjafari94@gmail.com">Niloufarjafari94@gmail.com</a></p>	<p><b>Objectives:</b> This study assessed the impact of rinsing water temperature on microleakage of dental composite restorations.</p> <p><b>Materials and Methods:</b> This in vitro study was conducted on 72 extracted premolars. Buccal and lingual Class V cavities were prepared with gingival margins 1mm beneath the cemento-enamel junction (CEJ) and occlusal margins 2mm above the CEJ. The specimens were classified into three groups (n=24) for rinsing with water at 23°C, 4°C, and 45°C before the bonding procedure. Lingual cavities were bonded with a self-etch adhesive, and buccal cavities were bonded with a total-etch adhesive. After composite restoration, the specimens underwent 1000 thermal cycles between 5-55°C. They were then divided into two groups for storage for either 24 hours or 2 months. Optical and electron microscopic assessments were performed to score microleakage at the occlusal and gingival margins of the specimens. The Kruskal-Wallis, Mann-Whitney, and Wilcoxon tests were run to analyze the data (alpha=0.05).</p> <p><b>Results:</b> Regardless of the bonding system and storage time, microleakage was significantly higher in the 4°C rinsing water group (P&lt;0.05). Gingival margins bonded with the total-etch adhesive had a significantly higher microleakage than the occlusal margins in both 23°C and 45°C groups (P&lt;0.05), but the self-etch adhesive showed no significant difference (P&gt;0.05).</p> <p><b>Conclusion:</b> According to the results of this in vitro study, rinsing the cavity with warm water (23°C and 45°C) appears to be an effective method to reduce the rate of microleakage.</p> <p><b>Keywords:</b> Adhesives; Composite Resins; Dental Leakage; Dentin; Temperature</p>

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### INTRODUCTION

Composite resins are extensively utilized in anterior and posterior dental restorations. However, their high technical sensitivity makes their actual clinical applications challenging [1,2]. Volumetric shrinkage is an inherent characteristic of these materials that causes stress generation at the tooth-restoration

interface, possibly causing microleakage [3]. Microleakage is a phenomenon defined as the passage of ions, molecules, bacteria, and liquids via the tooth-restoration interface. Its clinical manifestations include interfacial discoloration, tooth sensitivity, secondary caries, and sometimes pulpal pathologies [4,5]. Numerous techniques have been proposed to

prevent or minimize microleakage [6]. High bond strength is a crucial factor to reduce microleakage [7]. The adhesion mechanism of dentin is more complex than enamel, because of the presence of peritubular and intertubular dentin, high water content, and lower mineral elements than enamel, as well as a smear layer that penetrates into the superficial dentin during cavity preparation [8]. Bonding to dentin depends on the micromechanical adhesion of resin monomers between the collagen fibrils, forming both resin tags and a hybrid layer. The formation of a resin-dentin hybrid layer depends on smear layer modification and complete wetting of dentin surface by adhesive resin [9].

Several studies have shown that dentin bonding agents' temperature, dry air spray, restorative materials, and varying environmental temperatures affect the bond strength of dentin to composite resins [10-12]. Therefore, higher temperature of resin can improve wetting and conversion rate of resin monomers, which can consequently increase the bond strength and reduce microleakage during the restoration procedure [13,14], because heat increases the kinetic energy of molecules and can influence the properties of bonding agents and substrates [15,16]. Furthermore, a recent study showed that adhesives heated up to 55°C had higher final mechanical properties than those used at lower temperatures [17]. Heated composite resins have also been shown to have a lower microleakage rate than those immediately used after removal from a refrigerator [18].

Studies have shown that warm airflow within the thermotolerance zone (29°C to 56°C) of dentin has no pathological adverse effect on dentin, and dentin responds physiologically to this warm air blow [19,20]. The rinsing water temperature of dental unit may vary due to local weather conditions in different seasons of the year; thus, one strategy to increase the tooth surface temperature, which affects bonding and composite resin temperature, is to use warm water for tooth irrigation before bonding to boost the mechanical characteristics of the bonding interface [20]. Thus, the present study investigated the

impact of rinsing water temperature prior to the bonding procedure on microleakage of dental composite restorations. The null hypothesis was that the rinsing water temperature would have no significant effect on microleakage of restorations bonded with total-etch or self-etch adhesive systems.

## MATERIALS AND METHODS

A total of 72 human maxillary and mandibular premolars extracted for orthodontic purposes or periodontal disease were collected (ethical approval code: 389029). Prior to inclusion in the study, the teeth were inspected with dental loupes (Alltion Wuzhou Co., Guangxi, China) to ensure absence of caries, hypoplastic lesions, and cracks.

All debris, periodontal tissue residues, and calculi were eliminated from the tooth surface by an ultrasonic scaler, and the teeth were stored in 0.2% thymol solution at room temperature for 3 months before the experiments. The experimental design of the study was as follows: 72 teeth were randomly allocated to 3 groups (n=24) depending on the rinsing water temperature (4°C, 23°C, and 45°C). The 24 teeth allocated to each group received one buccal and one lingual standard Class V cavity preparation. The occluso-gingival dimension of the cavities was 3mm [2mm above the cemento-enamel junction (CEJ) and 1mm below the CEJ] (48 cavities). Buccal cavities were bonded using the total-etch technique. Lingual cavities were bonded by the self-etch technique (24 restorations per each bonding technique). After restoration and thermocycling, the teeth were stored for either 24 hours or 2 months, yielding 12 restorations for each experimental group (Fig. 1).

### **Cavity preparation:**

Two standard Class V cavities with a similar axial depth of 1.5mm at the enamel level and 1mm below the CEJ at the dentin level, occluso-gingival height of 3mm, and mesiodistal width of 3mm were prepared at the buccal and lingual surfaces of the teeth by a restorative faculty member using a barrel FG (ISO038) diamond bur (Diatiz CO, Tehran, Iran) and a high-speed handpiece under water and air spray.

To ensure optimal cutting efficiency, the bur was replaced after preparation of every four cavities. The teeth were randomly divided into three groups (Fig. 1) and stored in saline solution after the preparation of cavities and before the restorative procedures.

#### **Using water at different temperatures:**

To achieve 4°C (low-temperature) rinsing water, the irrigation syringes containing distilled water were placed in a refrigerator. To obtain 23°C (medium-temperature) and 45°C (high-temperature) rinsing water, the syringes containing distilled water were placed in a compound heater (UH-1 115V Vintage Dental Lab Utility Heater, Hanau, Germany). The water temperature in all three groups was verified by a Scan Temp485 thermometer (Dostmann Electronic GmbH, Wertheim, Germany) before the restorative procedures.

#### **Restorative procedures:**

All restorations were performed by the same operator. The buccal and lingual cavities were restored with Adper Single Bond (3M ESPE, MN, USA) total-etch system and Clearfil SE Bond self-etch bonding system, respectively. The teeth in all

groups were irrigated with water and dried with gentle airflow before restoration to remove debris. Table 1 presents the specifications of the adhesives and composite resin used in this study. The buccal cavity preparations were performed as follows: the enamel and dentin were etched with 37% phosphoric acid (Etch-Rite; Pulpdent Corporation, NY, USA) for 30 seconds and 15 seconds, respectively. The specimens were irrigated with water and air-sprayed for 10 seconds to remove the etchant. In this stage, 5-mL water syringes containing 4°C, 23°C, or 45°C distilled water were used to rinse the cavities again according to their group allocation. To prevent over-drying, excess water was removed with cotton pellets. The surface temperature was verified with a digital thermometer (Scan Temp485; Dostmann Electronic GmbH, Reicholzheim, Wertheim, Germany) with 0.94°C accuracy. In all groups, the procedure was continued if the tooth surface temperature was confirmed to be equal to the rinsing water temperature; otherwise, the surface was rinsed again with the respective water to reach the desired temperature.

Total	Premolars (n=72)											
Temperature (n=24)	4°C - LT				23°C - MT				45°C - HT			
Bonding* (n=24)	SE		TE		SE		TE		SE		TE	
Storage (n=12)	24h	2m	24h	2m	24h	2m	24h	2m	24h	2m	24h	2m

**Fig 1.** Study design illustrating the distribution of specimens among the groups. (\*2 cavity preparations per tooth: Buccal cavity was restored with total-etch adhesive and lingual cavity with self-etch adhesive).

**Table 1.** Description of materials used in the present study

Material description	Name/Manufacturer	Composition	Batch no.
<b>Total-Etch System</b>	Adper Single Bond (3M ESPE, MN, USA)	Adhesive: Bis-GMA, HEMA, dimethacrylate, polyalkanoic acid copolymer, initiators, water, and ethanol	N662648
<b>Self-Etch System</b>	Clearfil SE Bond (Kuraray co, Osaka, Japan)	Primer: MDP, HEMA, hydrophilic dimethacrylate, N, N diethanol p-toluidine, camphorquinone bond, silanated colloidal silica	9G0219
<b>Microhybrid Composite Resin</b>	Filtek Z250 (3M ESPE, MN, USA)	Organic matrix: BisGMA, UDMA, BisEMA; Camphorquinone (initiator); Filler: zirconia/Silica	N144264

Adper Single Bond was applied on the cavity surface in two layers according to the manufacturer's protocols. Each layer was applied with a microbrush for 15 seconds, and its solvent was evaporated with mild airflow from a distance of 10cm for an average of 5 seconds. Finally, the bonding agent was subjected to light curing for 20 seconds by Coltlux 75 (Coltene/Whaledent AG, Altstatten, Switzerland) light-curing unit with 1000mW/cm<sup>2</sup> power intensity. The cavity was restored with Filtek Z250 microhybrid composite resin (3M ESPE, MN, USA) using incremental application technique, and each increment underwent light curing for 40 seconds using the same device used for curing of the bonding agent. In the lingual cavities, the enamel margins were etched for 30 seconds and then irrigated with water at the desired temperature (4°C, 23°C, or 45°C) for 10 seconds as previously described. After ensuring the accuracy of the surface temperature, two coats of the self-etch primer (Kuraray Co., Osaka, Japan) were applied on the surface by a microbrush with slight agitation based on the manufacturer's protocols. After the solvent was evaporated with oil-free air spray from a distance of 10 cm for about 20 seconds, the adhesive (Kuraray Co., Osaka, Japan) was applied on the cavity by another microbrush and light-cured for 20 seconds. These cavities were also restored with microhybrid composite resin (3M ESPE, MN, USA) using the incremental technique, and each layer was light-cured for 40 seconds.

The restorations were finished by a high-speed fine diamond polishing bur (Teeskaavan, Tehran, Iran) and polishing discs (Opti; Kerr Dental, Bioggio, Switzerland), and were incubated in an incubator (Behdad, Tehran, Iran) at 37°C for 24 hours. The specimens were subjected to 1,000 thermal cycles at 5-55°C, with a dwell time of 30 seconds for aging and a transfer time of 15 seconds. Then, the specimens were stored for 24 hours and 2 months. The 24-hour specimens were tested, and the 2-month specimens were kept in the incubator for an

additional 60 days.

After storage, the root apices and the coronal portion were sealed with two layers of nail varnish up to 1mm around the restored cavities to stop dye penetration into the tooth and through the tooth-restoration interface. Next, the specimens were immersed in 0.5% Fuchsin solution (Atom Scientific Ltd Hyde, Cheshire, United Kingdom) for 24 hours and were then irrigated with distilled water for 2 minutes. Next, the specimens were mounted in self-cure acrylic resin (Acropars, Marlic Medical Industries Co., Tehran, Iran) and cut by a diamond bur (Proxxon Inc., Föhren, Germany) buccolingually from the center of the Class V restorations, parallel to the tooth longitudinal axis.

#### ***Microleakage analysis:***

The gingival and occlusal margins of all specimens were analyzed by a blind operator under an optical microscope (MGC-10N9116734, Ningbo Yongxin Optics Co., Ltd. Ningbo, Zhejiang Province, China) at 32× magnification. Two scores were reported for each of the occlusal and gingival margins in the buccal and lingual cavities. The microleakage was scored from 0 to 3 according to ISO/TS11405 (2003) [21] as follows:

**0:** No dye penetration

**1:** Dye penetration to ½ of the cavity depth

**2:** Dye penetration more than ½ of the cavity depth but not reaching the axial wall

**3:** Dye penetration through cavity depth to the axial wall

#### ***Preparation of specimens for scanning electron microscopic (SEM) analysis:***

A section was prepared from each specimen for SEM analysis to assess the formation of the hybrid layer and presence of any gaps or voids between the cavity margins and the restorative material. First, the specimens were demineralized in 6 N HCl solution for 8 hours, and placed in an ultrasonic cleaner (Esfahan Faraz Mehr Co, Isfahan, Iran) for 10 minutes. The specimens were then placed in 2.5% sodium hypochlorite (Jentex Alkalis, Chennai, India) for 10 minutes to eliminate the organic phase. Finally, they were placed in an ultrasonic water bath (Esfahan Faraz Mehr Co, Isfahan, Iran) for 10 minutes and polished

with coarse, medium, and fine paper discs (OptiDisc; Kerr Dental, Bioggio, Switzerland) under copious water irrigation in order to create a smooth, debris-free surface, which was confirmed through visual inspection. The specimens were then stored in a dry environment for 24 hours, and prepared for gold sputtering (BAL-TEC, CA, USA) prior to SEM analysis (SeronTechnology INC, Gyeonggi-do, Korea).

#### Statistical analysis:

SPSS version 23 (SPSS Inc., Chicago, IL, USA) was used for data analysis. Since the microleakage rate was rank-based, the Kruskal-Wallis non-parametric test was run. For between-group comparisons of independent groups (enamel and dentin margins in each group), the Mann-Whitney and Dunn Bonferroni tests were used. Furthermore, the Wilcoxon test was run for pairwise comparison of the groups based on the margin type and bonding system.  $P \leq 0.05$  was considered significant.

## RESULTS

### Marginal microleakage by ranking:

The Kruskal-Wallis test indicated the highest dye penetration in the 4°C rinsing water group, irrespective of storage time (24 hours or 2 months) and bonding method, in both occlusal and gingival margins ( $P < 0.000$  for the self-etch system in both the gingival and occlusal margins,  $P = 0.033$  for the gingival margins in the total-etch system, and  $P < 0.000$  for the occlusal margins in the total-etch system).

The results of comparison between different thermal and storage conditions in both gingival and occlusal margins within each bonding system are shown in Table 2.

Pairwise comparison of microleakage scores indicated no significant difference between the occlusal and gingival margins within each bonding technique and similar storage conditions when 4°C rinsing water was used (24-hour total-etch or self-etch, and 2-month total-etch or self-etch) ( $P > 0.05$ ). However, there was a significant difference between the gingival and occlusal margins in groups bonded with the total-etch technique and rinsed with 23°C and 45°C water ( $P < 0.05$ ).

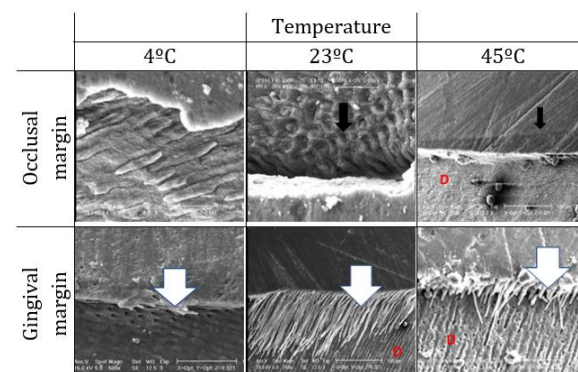
**Table 2.** Comparison of the two bonding systems in terms of microleakage

Groups	SE vs. TE Occlusal margin	SE vs. TE Gingival margin
MT 24h	0.414 <sup>1a</sup>	0.055 <sup>1b</sup>
LT 24h	0.18 <sup>2c</sup>	0.131 <sup>2c</sup>
HT 24h	0.317 <sup>1a</sup>	0.1 <sup>1b</sup>
MT 2m	0.084 <sup>1a</sup>	0.717 <sup>1b</sup>
LT 2m	0.435 <sup>2c</sup>	1 <sup>2c</sup>
HT 2m	0.272 <sup>1a</sup>	0.96 <sup>1b</sup>

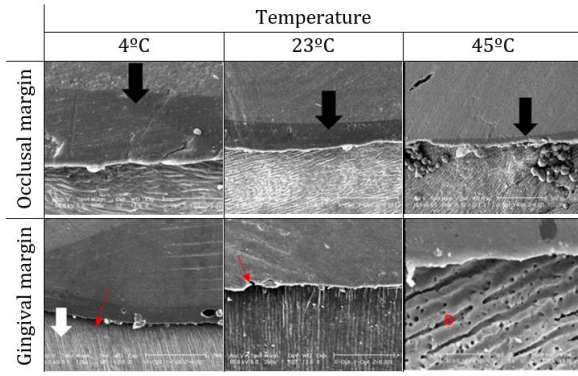
In rows, comparison of the same alphabetic letters showed no significant difference. In columns, the same superscripted numbers show no significant difference. SE: Self-etch, TE: Total-etch, LT: Low temperature, MT: Medium temperature, HT: High temperature, h: hours, m: months

### Morphological analysis of resin interaction with enamel and dentin:

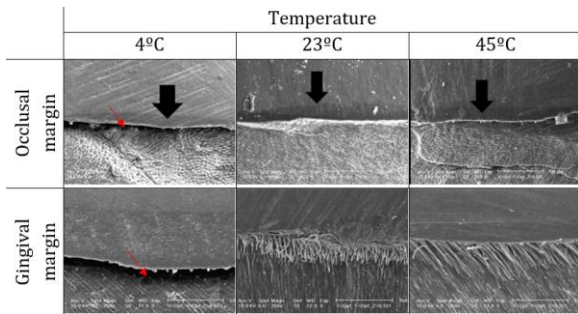
Figures 2-5 show the SEM micrographs of each group. The 24-hour total-etch group showed long resin tags combined with a dense hybrid layer. Some degradation of the hybrid layer was seen after two months of storage. When the self-etch system was applied, no resin tags were observed, while a distinct gap was observed between the adhesive resin and dentin substrate. The analysis of the morphology of the adhesive interface showed that the length and frequency of resin tags and the hybrid layer thickness at the gingival margins were the lowest at 4°C and the highest at 23°C. The thickness of the hybrid layer remained almost constant in both bonding



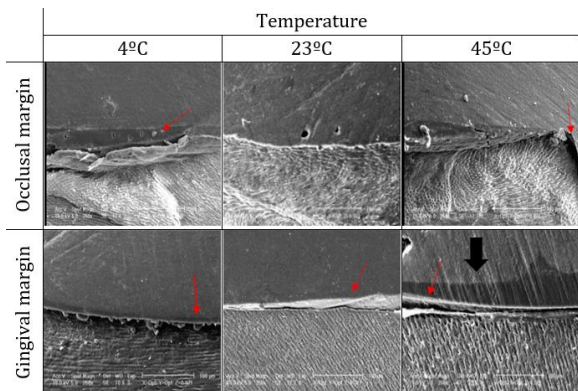
**Fig 2.** SEM micrographs of the dental interface in total etch group after using rinsing water at different temperatures following a 24-hour period (white arrow indicates resin tags; D -: dentin; Black arrow: adhesive layer; Red arrow: a gap between adhesive and dentin substrate).



**Fig 3.** SEM micrographs of the dental interface in self-etch group after using rinsing water at different temperatures following a 24-hour. (white arrow indicates resin tags; D -: dentin; Black arrow: adhesive layer; Red arrow: a gap between adhesive and dentin substrate).



**Fig 4.** SEM micrographs of the dental interface in the total-etch group after using rinsing water at different temperatures following a 2-month period. (Black arrow: adhesive layer; Red arrow: a gap between adhesive and dentin substrate).



**Fig 5.** SEM micrographs of the dental interface in the self-etch groups after using rinsing water at different temperatures following a 2-month period. (Black arrow: adhesive layer; Red arrow: a gap between adhesive and dentin substrate).

systems over 2 months at 23°C and 45°C. In both systems, the dye penetration depth was similar in the 23°C and 45°C groups and lower than that in the 4°C group.

### DISCUSSION

The study results indicated the impact of rinsing water temperature on microleakage at the dentin bonding agent and dental substrate interface. Microleakage was significantly higher in the 4°C group than the 23°C and 45°C groups in both bonding systems and both occlusal and gingival margins. Hence, the null hypothesis, which stated that rinsing water temperature would have no significant effect on the microleakage rate at the dentin/adhesive interface, was rejected.

Class V cavities were investigated in this study since they have easy restoration and do not need macro-mechanical undercuts during cavity preparation. Therefore, the sealing ability of composite resins is judged solely based on their bonding potential [22]. Also, both the enamel and dentin margins can be assessed in this type of cavity. Restoring cervical lesions can be difficult because their margin is partly in the enamel and partly in dentin or cementum [23].

Temperature rise of the substrate may enhance the temperature of the composite resin and adhesive. Therefore, one reason for the reduced microleakage at higher rinsing water temperatures at both 24 hours and 2 months could be improved adhesive properties, i.e. easier evaporation of the solvent, increased conversion rate of monomers, and reduced water sorption and adhesive solubility in the long run [24]. It has been suggested that all mechanical properties of adhesives can be improved by an increase in temperature [17]. Studies have shown that increased temperature decreases adhesive viscosity and enhances the wetting of enamel and dentin surfaces [14,25]. Increased temperature also improves the mechanical properties of composite resins and facilitates their application [26].

However, the impact of temperature on the properties of composite resin restorations is controversial. A new study showed that

composite preheating is a worthwhile method to enhance its adaptability and reduce the microleakage of composite restorations, thereby decreasing the microleakage rate [27]. However, another study reported that temperature increase did not affect the mechanical characteristics of adhesive [28]. Some others indicated that increasing the temperature of adhesives only improved the bond strength of etch-and-rinse bonding systems and not self-etch adhesives [20,29]. This difference can be due to the use of shear bond strength test in these studies and failure to evaluate other mechanical features of adhesives.

In the etch-and-rinse systems, the collagen fibers collapse following etching, as a result of slight removal of inorganic content, and interpeptide hydrogen bonds start to form when collagen fibers approach one another [30], causing more contraction of the collagen network. The strength of hydrogen bonds increases with a decrease in temperature [31]. Therefore, the collagen network, unlike when it is rinsed with warmer water, is not contracted by the primers containing HEMA and solvent, and the breakage of hydrogen bonds and penetration of primers become more difficult. On the other hand, the free energy of the dentin and enamel surfaces is reduced with temperature loss; thus, the contact angle between the surface and liquid increases and liquid is not spread over the surface and is less penetrated [31,32]. Lack of a significant difference in microleakage of cavities rinsed with 23°C and 45°C water can be due to the surface energy required for overcoming the surface tension of resin at 23°C; thus, higher temperatures that decrease viscosity may not be associated with a stronger bond [33].

As expected, the microleakage rate was higher in the gingival margin than in the occlusal margin in the etch-and-rinse bonding system when 23°C and 45°C rinsing water temperatures were used, and these results are the same as those obtained by Filtek Z250 in a study by Madani et al [34].

However, the microleakage rate was similar between the occlusal and gingival margins at

both storage times when 4°C water was used for rinsing, probably because of the higher overall microleakage rate. Comparison of microleakage at the occlusal margins of cavities treated with the total-etch and self-etch systems showed no significant effect of the bonding system in similar margins which may be due to the same etching protocol (37% phosphoric acid for 30 seconds) related to the same resin penetration after the bonding procedure [35].

There was also no significant difference in the microleakage rate between the 24-hour and 2-month periods in all experimental groups in spite of the degradation of the interface when the morphology of the adhesive interface was analyzed. Similarly, another study indicated no difference after the thermocycling and storage periods [36].

Moreover, the results revealed a higher microleakage rate when the substrate was irrigated with 4°C water than with 23°C and 45°C water, which affected the temperature of the substrate, its surface energy, and possibly the temperature of the adhesive and composite applied on the substrate, even though these properties were not explicitly measured. The results obtained for dye penetration in this study were confirmed by SEM analysis. Thus, during the cold season when the temperature of the rinsing water may be low, the water should be kept at room temperature for a few minutes before use to make its temperature closer to 23°C.

Further research is required to accurately measure the temperature of each restorative material after deliberately changing the temperature of the substrate. One limitation of this study was the failure to investigate the impact of water temperature on the microleakage over a longer period of thermocycling (e.g. 6 months). Hence, it would be helpful to conduct further studies to consider the above points and compare the results.

## CONCLUSION

The findings of this study showed that using 4°C water for rinsing can remarkably increase the microleakage rate of occlusal and gingival margins of composite restorations. Hence, clinicians must avoid tooth surface irrigation

with low-temperature water. In cold seasons, water should be kept at room temperature (~23 °C) for a few minutes before use to boost the clinical performance of composite restorations.

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#### CONFLICT OF INTEREST STATEMENT

None declared.

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