

## EFFECT OF LINERS ON COMPOSITE RESIN MICROLEAKAGE AFTER SELECTIVE CARIOUS LESION REMOVAL. AN EXPERIMENTAL STUDY.

Efecto de los recubrimiento en la microfiltración de resina compuesta después de la eliminación selectiva de lesiones cariosas. Un estudio experimental.

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### ABSTRACT:

**Objective:** To evaluate microleakage of composite resins (CR) placed over different cavitary liners after managing deep caries lesions through selective removal of soft carious tissue to soft dentin (SRCT-S).

**Material and Methods:** Fifty four human teeth were collected for microleakage testing. Each assay comprised ICDAS 5 or ICDAS 6 carious lesions and sound teeth for controls. Sound teeth were prepared with cavities that mirrored the carious teeth cavities, which were prepared with SRCT-S. Sound and carious teeth were further randomly assigned to one of the three experimental groups: Group A: universal adhesive (UA) + CR, Group B: glass ionomer cement liner + UA + CR, and Group C: calcium hydroxide + UA + CR. Occlusal microleakage (OM) and cervical microleakage (CM) was classified within one of 5 depth categories. ANOVA and Chi-square tests were computed ( $p < 0.05$ ).

**Results:** OM and CM were similarly distributed across subgroups ( $p > 0.05$ ). All Group C samples with carious lesions presented some degree of microleakage. However, no statistically significant differences were observed between groups and within each group ( $p > 0.05$ ).

**Conclusion:** Teeth restored with CR after SRCT-S using calcium hydroxide as a liner material seem to exhibit higher microleakage than those restored using glass ionomer or UA alone. Further clinical research is needed to deepen into these findings.

**Clinical significance:** The application of calcium hydroxide as a liner under a composite resin may reduce the longevity of a restoration after performing selective or partial removal of carious tissues. Clinicians should rethink the need of using calcium hydroxide for this application, albeit limited of clinical evidence.

### KEYWORDS:

*Dental cavity lining; Adhesion; Dental caries; Composite resins; Glass Ionomer Cements; Microleakage.*

## RESUMEN:

**Objetivo:** Evaluar la microfiltración de resinas compuestas (RC) colocadas sobre diferentes liners cavitarios después del manejo de lesiones de caries profundas mediante la remoción selectiva de tejido cariado blando hasta dentina blanda (SRCT-S).

**Material y Métodos:** Se recolectaron 54 dientes humanos para pruebas de microfiltración. Cada ensayo comprendía lesiones cariosas ICDAS 5 o ICDAS 6 y dientes sanos para los controles. Se prepararon dientes sanos con cavidades que reflejaban las cavidades de los dientes cariados, que se prepararon con SRCT-S. Los dientes sanos y cariados se asignaron al azar a uno de los tres grupos experimentales: Grupo A: adhesivo universal (AU) + RC, Grupo B: revestimiento de cemento de ionómero de vidrio + AU + RC, y Grupo C: hidróxido de calcio + AU+ RC. La microfiltración oclusal (MO) y la microfiltración cervical (MC) se clasificaron dentro de una de las 5 categorías de profundidad. Se calcularon las pruebas ANOVA y Chi-cuadrado ( $p < 0,05$ ).

**Resultados:** La MO y MC se distribuyeron de manera similar en los subgrupos ( $p > 0,05$ ). Todas las muestras del Grupo C con

lesiones cariosas presentaron algún grado de microfiltración. Sin embargo, no se observaron diferencias estadísticamente significativas entre grupos y dentro de cada grupo ( $p > 0,05$ ).

**Conclusión:** Los dientes restaurados con RC después de SRCT-S usando hidróxido de calcio como material de revestimiento parecen exhibir una mayor microfiltración que aquellos restaurados usando ionómero de vidrio o AU solo. Se necesita más investigación clínica para profundizar estos hallazgos.

**Relevancia clínica:** la aplicación de hidróxido de calcio como revestimiento debajo de una resina compuesta puede reducir la longevidad de una restauración después de realizar la eliminación selectiva o parcial de los tejidos cariados. Los médicos deberían reconsiderar la necesidad de usar hidróxido de calcio para esta aplicación, aunque no haya evidencia clínica.

## PALABRAS CLAVE:

*Recubrimiento de la Cavidad Dental; Adhesión; Resinas compuestas; Caries dental; Cementos de ionómero vítreo; Microfiltración*

## INTRODUCTION.

The Minimal Intervention Dentistry (MID) philosophy has meant an important change on the management of dental caries. This approach has incorporated non- and minimally invasive techniques, challenging the traditional restorative approach.<sup>1-3</sup> Once a minimally invasive restoration is indicated, there exists a range of restorative dental biomaterials, with biocompatible properties, to assist the recovery of form and function of affected teeth, maintaining the integrity of the dental tissues and minimizing tooth structure loss. Indeed, recent conservative therapies have shown to reduce pulpal exposure during carious tissue removal, maintaining pulpal vitality for longer periods of time.<sup>2,4</sup>

Unlike traditional complete removal, global current consensus recommend the selective removal of carious tissue to soft dentine (SRCT-S),

also called partial removal, for deep carious lesions in vital teeth.<sup>2,5</sup> SRCT-S maintains some carious tissue on the pulpal wall, while removing the peripheral affected dentin to sound tissue, avoiding pulp exposure, but ensuring optimal adhesion to the lateral walls.<sup>6</sup> This technique usually includes liners and the same-session restoration with permanent materials, mainly composite resin. Although certain liners seem capable of inducing tertiary dentin production and reducing pulpal inflammation,<sup>7</sup> evidence is still scarce.<sup>8-11</sup> Indeed, a systematic review reported that the evidence on this issue is still weak and more high-quality randomized controlled trials are needed.<sup>12</sup>

The most frequently used liners for the treatment of deep carious lesions are calcium hydroxide (CH), glass ionomer cements (GIC) and universal adhesives. CH has been traditionally used as a

liner to preserve pulp vitality for the management of deep carious lesions, including as direct pulp capping.<sup>13</sup>

CH has been used for many years, arguably due to its high pH, antibacterial properties and its capacity to induce calcified tissue formation through the release of calcium ions.<sup>14</sup> Nevertheless, CH cements have limited adhesion to dentine, high solubility, and low mechanical properties.

These limitations generate increased porosity (*i.e.* tunnel defects) and microleakage.<sup>15</sup> On the other hand, GIC are biocompatible materials with chemical adhesion to dental tissues with the capacity of releasing fluoride, low solubility, and able to provide an appropriate dentine seal.<sup>16</sup> It has been reported that GIC help in carious dentine remineralization through fluoride and strontium release.<sup>17</sup> Universal adhesives, also called eighth generation adhesives can be used as a self-conditioning, a total etch or a selective etch system.

They are characterized by the presence of acidic monomers that do not require a separate etching and rinsing steps in dentine. These adhesives are becoming popular due to the less technique-sensitive application and reduced clinical steps.<sup>18</sup> These systems contain acidic monomers that condition, demineralize and infiltrate enamel and dentine, simultaneously. The smear layer is not removed, and rinsing is not indicated when self-conditioning or selective etch techniques are performed. When etching and rinsing are not performed, the risk of over-conditioning dentine is reduced, minimizing adhesive monomer penetration, decreasing dentine permeability and post-operative sensitivity.<sup>19</sup> Self-etching adhesives have demonstrated adequate and stable chemical and micromechanical bond strength to dentine, even better bonding than etch and rinse systems.<sup>20</sup>

During the carious lesion removal process, remnant intertubular dentine is irregular and has more mineral content than normal dentine.<sup>21</sup> This can affect the tooth mechanical stability,<sup>11,21,22</sup> which is closely related with the marginal integrity of the restoration. During mastication, cusp flexion

increases, accelerating marginal deterioration and leading to microleakage with the eventual formation of secondary caries.<sup>23</sup> Conflicting results have shown that selective caries removal decreases fracture resistance in molars<sup>24</sup> and that this technique could not demonstrate reduction in fracture resistance or deficient marginal integrity.<sup>11,23</sup> Although composite resins seem to be the standard of care in restoring teeth with deep dentine lesions<sup>2,25</sup> bonding properties to carious tissues remain elusive and a matter of intense debate in restorative dentistry and cariology. Indeed, failure of resin restorations depends on the depth of the cavities and the restorative techniques.<sup>26</sup> There is a need to investigate the adhesive behavior of composite resins in conservative therapies, such as SRCT-S. Given the lack of robust evidence on the effects of SRCT-S and the need for lining materials, more research with clinically relevant experiments appears needed. "Therefore, the present study aimed at evaluating composite resin microleakage in restorations of deep caries lesions using different lining strategies over dental tissues managed with SRCT-S and restored with composite resin, *in vitro*."

## MATERIALS AND METHODS.

Approval to collect extracted tooth samples from the Scientific Ethics Committee of the University of Talca (2018013) was obtained. The sample comprised 54 human teeth, 27 with deep caries lesions, coded as ICDAS 5 or ICDAS 6, and 27 caries lesion-free and developmental defects-free (sound teeth). The cleaning of each tooth was performed using a white bristle prophylaxis brush with a water and fine pumice paste. The cleaned teeth were immersed into a 2% chlorhexidine solution for 10 minutes and preserved in 0.09% saline solution at room temperature until their preparation.

Caries teeth (n=27) and sound teeth (n=27) were assigned to microleakage tests. The study groups were categorized according to three different restoration techniques:

### Group A

Universal Adhesive (UA) + composite resin (CR),

Group B: GIC+UA+CR, and Group C: CH+UA+CR. For the microleakage tests, two subgroups of carious (n=9) and sound teeth (n=9) were selected for each study group (n=18 per group) (Figure 1A).

### Microleakage tests

Cavity preparation: A class II cavity was prepared in all specimens with caries lesions. The carious tissue was eliminated until reaching the soft dentin at the bottom of the cavity (slight resistance to the bur with minimal pressure). The carious lesion was eliminated completely at the walls of the cavity. Thus, lateral walls and marginal limits of the cavity were placed on sound enamel and dentin. Cavity opening was performed with a high-speed turbine (NSK-OBG10276 at 40,000 rpm), a round diamond bur (12 Fine grain - 545614) and a round carbide bur (811671) to remove carious dentin from the pulpal wall, keeping internal angles rounded.

Both burs were used with abundant cooling with water, and replaced after preparing 5 cavities with new burs. Manual removal was also done, to reach the deepest area and to have a tactile feeling of the soft tissue with manual excavators (Maillefer 63/64-71/72). All cavities were made by a single previously trained and calibrated operator. Also, the 27 homologous without carious lesions specimens were prepared, mirroring the size and shape of the cavities obtained from the carious teeth (Figure 2A). Subsequently, the specimen duos were randomly assigned to each study group and subgroup, as described in Figure 1.

### Treatments

Samples were treated with 3 different lining approaches, and all restored with composite resin. Thus, Group A was treated with an universal adhesive, Group B with a glass ionomer cement and Group C with calcium hydroxide, as described below:

#### Group A (UA)

Samples were selectively etched (only enamel) with 35% orthophosphoric acid for 15 s (Ultradent, UTAH, USA). After washing and drying the surface, water spray was applied to the occlusal surface for 15 seconds. Dry for 30 seconds with dry air.

Single Bond Universal Adhesive (3M Oral Care, Saint Paul, MN, USA) was applied with a disposable applicator covering the tooth surface and rubbing for 20 seconds, then blowing the liquid for 5 seconds with soft air, until it no longer moves and the solvent has completely evaporated. Polymerize the adhesive for 10 seconds with a curing light. and light cured for 10 seconds at a distance as close to the tooth and to the site of polymerization.

A LED light-curing lamp (Woodpecker P110 80024B) was used. For the filling, direct resin (3M ESPE FILTEK Z350 XT, LOT N924647-N899733) was used using the incremental technique. Four to 5 increments were applied, depending on the size of the cavity. The same number of increments was used for the experimental and control sample, standardized at 2 mm thickness and light-cured for 20 seconds per increment.

#### Group B (GIC)

Ketac Molar Easymix cement (3M ESPE, Seefeld, Germany) was prepared manually according to the manufacturer's instructions and applied as a liner at the bottom covering only the cavity floor. Subsequently, selective etching (enamel) with 35% orthophosphoric acid was performed. Single Bond Universal Adhesive and direct resin were applied similarly to Group A.

#### Group C (CH)

Calcium hydroxide cavity base Dycal® (Dentsply, Milford, DE, USA) was applied according to the manufacturer's instructions. The Same restorative technique (*i.e.*, etching, adhesive and direct resin) used in Group A y B was performed to fill the carious and sound tooth samples. All restored teeth were stored in a saline solution at room temperature for 24 hours. Restorations were finished and polished with Sof-Lex™ discs (3M Oral Care, Saint Paul, MN, USA).

After fixing the samples in methacrylate cylinders, 250 cycles of 10 kg axial loads were applied at 40 psi for 0.5 seconds in each cycle. Samples were exposed to 250 thermal cycles (20 seconds at 55°C and 20 seconds at 5°C, in water). Then, samples were immersed for 2 days in a

0.2% aqueous solution of methylene blue (Merck, Darmstadt, Germany). Finally, samples were cut using a handpiece and a fine-cut carborundum disk to separate the root from the crown. The crown was hemisected throughout the center of the restoration from mesial to distal (Figure 2B). One of the halves was selected for further analyses, based on the more severe microleakage score.

### Microleakage measures

An expert examiner, previously trained (85% kappa value) and blind for the restorative technique used, assessed microleakage. Penetration of the staining solution at the tooth-restoration interface was examined at the occlusal and cervical margins, using a light microscope at 10x magnification.

The following criteria were used to categorize the staining penetration (Figure 2B):

0: No distinct microleakage.

1: Slight microleakage. The dye penetrates less than the outer half of the gingival or the axial wall of the cavity.

2: Moderate microleakage. The dye diffuses beyond half of the gingival wall but does not reach the axio-pulpal wall in the proximal box. In the occlusal box, the dye extends past half of the axial wall without reaching the pulpal wall.

3: Major microleakage. The dye involves less than half of the axio-pulpal wall of the proximal box and

less than half of the pulpal wall of the occlusal box.

4: Severe microleakage. The dye extends over half the axio-pulpal wall of the proximal box and over half the pulpal wall of the occlusal box.

### Statistical analysis

Occlusal microleakage (OM) and cervical microleakage (CM) were analyzed separately. The proportion of each microleakage score was compared by group and subgroup samples using Chi-squared ( $\chi^2$ ) tests. A  $p$ -value  $<0.05$  was considered significant. Data analyses were conducted using SPSS v.24 (IBM, NY, USA)

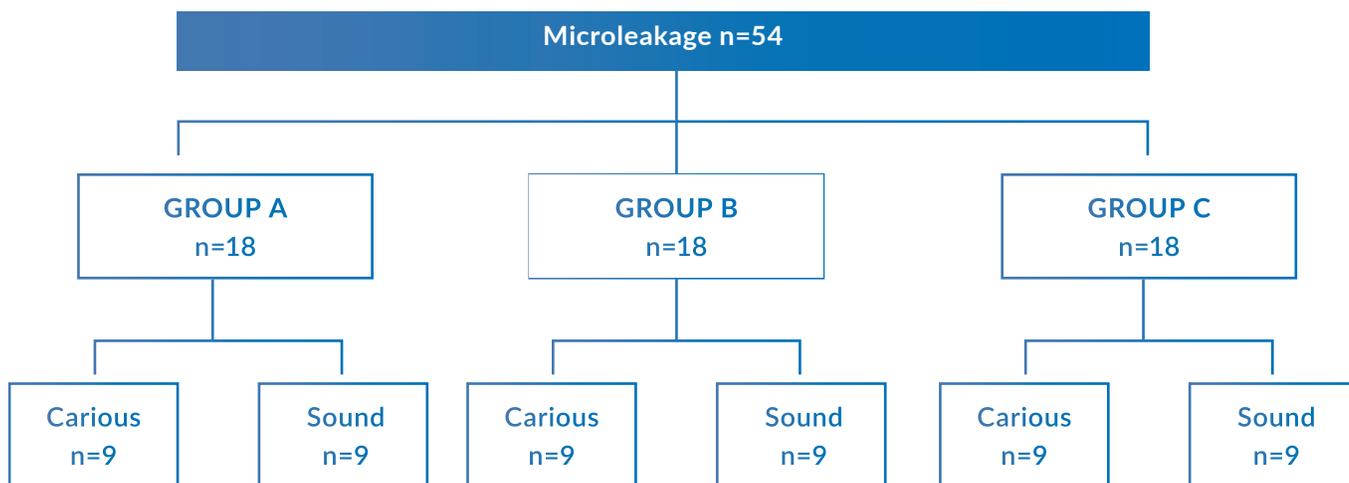
## RESULTS.

Occlusal microleakage scores distributed similarly across carious and sound subgroups within each experimental group (A, B or C). All Group C samples with carious lesions presented some degree of microleakage.

However, no statistically significant differences were observed among groups and within each group ( $p>0.05$ ).

Percentage distributions for the microleakage category of each subgroup is represented in Figure 3A. Comparable results were found regarding CM, without statistically significant differences across the samples ( $p>0.05$ ). Only one sound tooth from Group B showed no CM (Figure 3B).

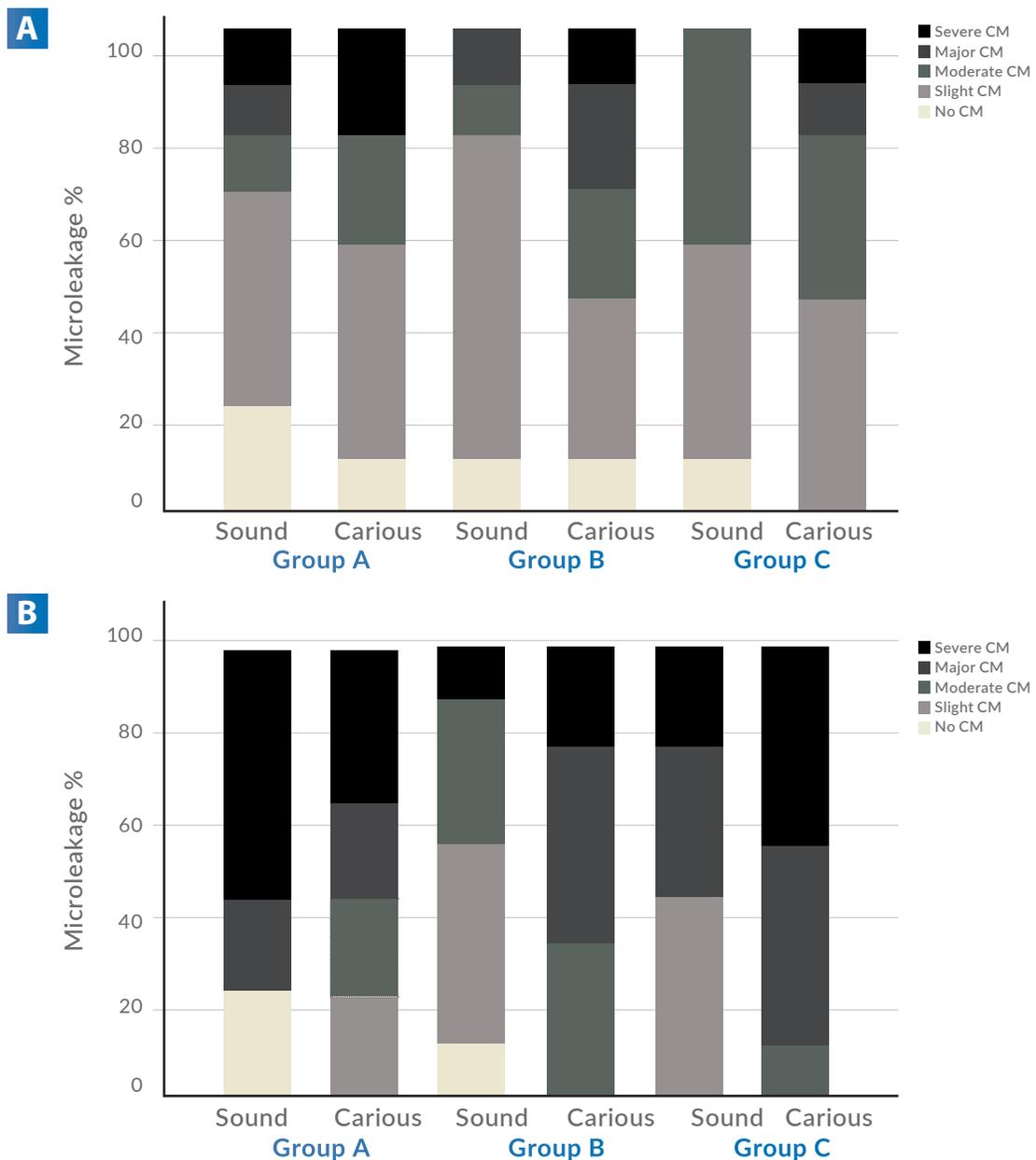
Figure 1. Sample distribution for Microleakage Testing.



**Figure 2.** Image (A) shows an example of a cavity preparation in specimens with carious lesions (I) and without carious lesions (II). Panel (B) shows the microleakage scores for the occlusal and proximal boxes of the cavities.



**Figure 3.** Percentage distribution of OM (A) and CM (B) by experimental subgroups and the presence of carious tissue. Bars indicate the percentage of microleakage according to the severity of the staining, as indicated by color coding.



## DISCUSSION.

Although there is a consensus on the need to perform partial removal of dental tissues during tissue excavation for deep caries lesions in order to avoid pulp exposure and preserve tooth structure<sup>2</sup>, evidence is still limited, and no studies have shown the need to use a pulp protection prior to composite resin placement or whether one type is better than another.

This study experimentally demonstrated that there were no differences in microleakage levels between the liners. However, CH represented the most severe levels for CM and OM.

Current dental materials have evolved, improving their properties and performance, focusing on making clinical procedures simpler, with better results in less time. Composite resins and universal adhesive systems are the preferred dental material when carious teeth are managed with SRCT-S.<sup>25</sup> Universal dental adhesives were incorporated as versatile and multifunctional systems with less application steps and they are compatible with all dental tissue treatment modalities. These systems are also capable of bonding with diverse restorative materials used in combination with adequate surface treatments.<sup>27,28</sup> The bonding characteristics of adhesive systems are essential to achieve the long-term clinical success of resin restorations. Sealing restoration margins protects against microleakage and prevents following complications such as post-operative sensitivity, marginal discoloration and secondary caries lesions development.<sup>22</sup> The marginal Integrity and secondary caries lesions are often considered as the main reason for restoration failure. However, clinical trials have not confirmed this hypothesis.<sup>29,30</sup>

The present study results demonstrated that OM and CM behaved similarly in all the study groups, both over carious lesions and sound tissues, although cervical margins presented a higher percentage of severe microleakage compared to occlusal areas. Reasons for this could be related to the etching and the bond strength of resin restorations, which perform better in occlusal enamel due to its width,

histologic structure, and quantity.<sup>31,32</sup> Another study supports that higher CM is not related with the liner characteristics but with the thicker enamel width found in cervical areas. This could result in a bonding reduction of the composite resin to the dental structure.<sup>33</sup>

Although no statistically significant differences were found, data showed higher microleakage when CH was used, with 100% of the CH-samples exhibiting some degree of OM and CM. A previous study using CH determined that 50% of the samples had some degree of microleakage.<sup>34</sup> Nevertheless, CH has been used for decades as a liner due to its chemical and biological properties<sup>35</sup>, in spite of its high solubility and lack of mechanical and adhesive properties.<sup>36,37</sup>

On the other hand, it is important to consider that polymerization shrinkage is an inherent characteristic of composite resins; therefore, a bulkier composite resin restoration could lead to increased microleakage. Furthermore, adhesive systems have some limitations that can explain the results during adhesive and microleakage tests. For instance, the thin hybrid-layer that self-etch adhesive produces on dentine, less than 10 µm thick, may not be resistant enough to withstand polymerization shrinkage stress.<sup>38</sup> Also, an incomplete polymerization, the thicker volume of composite resin, may favor water sorption and affect mechanical and chemical properties.<sup>38</sup>

A vast number of universal adhesives have the monofunctional monomer HEMA. This is a highly hydrophilic small molecule that works as a diffusion agent, that can promote water sorption from the dentine into the adhesive interface, making it susceptible to hydrolytic degradation.<sup>38</sup> Finally, the incorporation of silane into the adhesive system decreases acidity in the solution (pH>2.5), limiting dentine self-conditioning and bonding efficacy.<sup>39,40</sup>

Importantly, these findings showed comparable microleakage between carious lesions and sound enamel, suggesting that microleakage is not related to the nature of the adhering tissues, but it may be the result of the combined effect of the

polymerization contraction and lesion depth.<sup>23</sup>

Explanations for these findings may also arise from the high solubility of CH. Mount *et al.*,<sup>41</sup> reported similar results, whereby CH showed higher microleakage compared with resin-modified GIC and composite resins, discouraging the use of CH as a liner for composite resin restorations.<sup>41</sup> Similarly, we failed to detect significance between GIC and self-etching adhesive used as liners. There is some evidence, however, showing that composite resin had higher likelihood of fracture when GIC was used as liner instead of total etching technique.<sup>42</sup> The application of a liner under a composite resin may be considered a predictor for significant reductions of restoration longevity. The rationale for this assertion is that the lack of adhesion of the liner to the dental structure creates unfavorable mechanical behavior.<sup>43</sup>

On the other hand, a consideration for the use of liners in class I and II composite resin restorations is post-operative sensitivity. It has been argued that liners could counteract this clinical complication, but there is only weak evidence to support this statement.<sup>44</sup> Composite resins have an insulating effect. Thus, the need for the use of a liner may be questionable, as the restorative material may provide enough thermal insulation. As discussed above, using a liner limits the available surface for bonding, but reduces the thickness of the composite resin, which offers better mechanical properties to the restoration. Moreover, the application of an adhesive agent in the remnant carious dentine has a relevant sealing effect protecting the pulp from many stimuli and bacterial penetration. Consistent with our findings, the indication of a liner under the composite resin performed over a SRCT-S seems unnecessary.<sup>45</sup>

Studies focusing on biomaterials, adhesion and their interaction with the biological tissues are concentrating the interest of the international dental research community. Yet, we acknowledge the inherent limitations of an *in vitro* study. Despite the efforts to mirror the caries with the healthy samples, there might be configurational differences. Also, teeth come from different patients and the tissue may have certain variations among the donors. Lastly, these findings may not necessarily be reproduced under clinical conditions, as the sample size was minor, where other variables concur and may affect the final outcomes. There is a need for further longitudinal clinical trials to draw more evidence-based conclusions about the advantage of using or not liners to protect the pulpal tissue when selective removal of carious tissues is performed.

## CONCLUSION.

Composite restorations carried out after SRCT-S seem to produce similar microleakage values than those performed over mirrored cavities on sound tissues. No differences were detected between UA and GIC, applied either on sound or carious tissues. CH used after SRCT-S appears to induce an unfavorable adhesive behavior of the restoration. Further clinical studies are necessary to confirm these experimental findings.

#### **Conflict of interests:**

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

#### **Ethics approval:**

Study was approved by the Scientific Ethics Committee of the University of Talca (2018013).

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#### **Authors' contributions:**

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