











Evaluation of marginal infiltration by varying the adhesive protocol: *In Vitro* study

Avaliação da infiltração marginal pela variação do protocolo adesivo: estudo In Vitro

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Abstract

Aim: The aim of this study was to evaluate the sealing of composite resin restorations, observing marginal infiltration through a conventional resin by varying the adhesive technique.

Materials and Methods: Cavity preparations were performed on the buccal and lingual surfaces with margins at the cement-enamel junction. The samples were divided into three groups (n=10): Group 1-Selective conditioning technique; Group 2-Total conditioning technique; Group 3-Non-conditioning. Then, the restorations were made using conventional composite resin, immersed in methylene blue (C16H18ClN3S) for seven days and evaluated in a standardized manner, through scores, and analyzed the penetration depth of the dye.

Results: The data were submitted to the Tukey test with a significance level of 5%. The results showed that the samples that were not conditioned obtained the highest infiltration rates (p=0.0001).

Discussion: The integrity of the margins of a restoration is essential to clinical success, since its absence is directly related to the emergence of interface infiltrations.

Conclusion: The authors concluded that the worst group was the one without etching; enamel showed less infiltration than dentin for all groups; the technique of selective etching and total etching obtained similar results regarding microleakage.

Keywords

Composite Resins, Dentin, Dental Leakage

Resumo

Objetivo: O objetivo deste estudo foi avaliar a vedação de restaurações de resina composta, observando infiltração marginal através de uma resina convencional variando a técnica adesiva.

Materiais e Métodos: Os preparos cavitários foram realizados nas faces vestibular e lingual com margens na junção cimento-esmalte. As amostras foram divididas em três grupos (n=10): Grupo 1 - Técnica de condicionamento seletivo; Grupo 2 - Técnica de condicionamento total; Grupo 3 - Não condicionamento. Em seguida, as restaurações foram feitas com resina composta convencional, imersa em azul de metileno (C16H18ClN3S) por sete dias e avaliada de forma padronizada, por meio de escores, e analisada a profundidade de penetração do corante.

Resultados: Os dados foram submetidos ao teste de Tukey com nível de significância de 5%. Os resultados mostraram que as amostras não condicionadas obtiveram as maiores taxas de infiltração (p=0,0001).

Discussão: A integridade das margens de uma restauração é essencial para o sucesso clínico, uma vez que sua ausência está diretamente relacionada ao surgimento de infiltrações na interface.

Conclusão: Os autores concluíram que o pior grupo foi o sem condicionamento, o esmalte apresentou menor infiltração que a dentina em todos os grupos, a técnica de condicionamento ácido seletivo e o condicionamento total obtiveram resultados semelhantes quanto ao microinfiltração.

Palavras-chave

Resinas compostas, Dentina, Infiltração dentária

1 INTRODUCTION

The introductory framework of “adhesive dentistry” was the acid conditioning technique described by Buonocore in 1955, which proposed that the use of phosphoric acid on the enamel surface formed microporosities that allowed the bonding between restorative material and dental substrate (Buonocore, 1955). Adhesive procedures consist of replacing lost minerals (enamel and dentin) with resin monomers that are incorporated into the dental structure (Muñoz *et al.*, 2013). Due to differences between the biological structures of enamel and dentin, the adhesion occurs differently between these tissues. Due to a complex histological structure, dentin has greater sensitivity to the adhesive technique employed (Swift; Perdigão; Heymann, 1995). Dentin adhesion occurs through the formation of the hybrid layer. Described by Nakabayashi in 1982, this layer results from the interaction of dentin collagen matrix with resin infiltration (Nakabayashi; Kojima; Masuhara, 1982). The universal adhesive can be used through different technical features, such as: conventional technique and self-conditioning technique. The conventional technique recommends acid attack to expose collagen fibrils for subsequent penetration of monomers into the interfibrillary spaces. The self-conditioning technique uses the treatment of dentin through an acidic primer that demineralizes the tissue and concomitantly allows the infiltration of the adhesive (Van Meerbeek *et al.*, 1998). Demineralized and non-infiltrated areas by adhesive are subject to infiltration, a process that can be observed through the staining technique (Carvalho *et al.*, 2005). This process is defined as being “the flow of oral fluid and bacteria in the microscopic gap, between the surface of a prepared tooth and a restorative material (Anusavice, 2005). By invading the dentinal limits of the resin, spaces and porosities are generated in the hybrid layer, deteriorating the binding area, and causing dentin sensitivity, secondary caries, and pulp inflammation (Bail *et al.*, 2012). This study aims to evaluate the effect of marginal infiltration and sealing capacity in conventional composite resin restorations using universal adhesive system in conventional and self-conditioning mode.

2 MATERIALS AND METHODS

Ten healthy teeth were submitted to the Research Ethics Committee with number CAAE 39698420.3.0000.5237. The teeth were cleaned and stored in distilled water until the moment of the experiment.

2.1 Specimen Preparation

In the teeth, standardized cavities were prepared, with 1 cavity on the vestibular surface and 2 in the palatine/lingual (figure 1) with diamond drills no. 1090 and n°1014 (KG Sorensen-Brazil) in high rotation (Kavo-Germany), under constant cooling. The drills were changed after every 5 prepares. The cavities followed the following dimensions: 2 mm deep, 2 mm of mesiodistal extension and 2 mm of occluso cervical extension and with gingival margins located 1 mm below the JCE. The dimensions of each cavity were confirmed with a digital caliper. Immediately after preparation, the specimens were randomly divided into 3 groups (10 cavities per group), in which each group presented the interface enamel(E) and dentin(D) according to the conditioning method, using the self-conditioning, conventional and acid-free adhesive technique prior. After preparation, the adhesive protocol was performed with the Universal Adhesive System (AmbarAPS-FGM-Brazil), relative to each group and the cavities were restored with the conventional composite resin (VittraAPS-FGM-Brazil). The conventional resin was inserted in two increments, with photoactivation by 20 seconds at each increment with the light curing (Valo Cordless-Ultradent-Brazil). The restorations were finished with thin-grained diamond tips no. 3118F (KG Sorensen-Brazil) in high rotation, with air/water irrigation, and polishing was performed with discs of different granulations (Diamond FGM-Brazil) and silicon carbide bowls (American Burrs-Brazil).

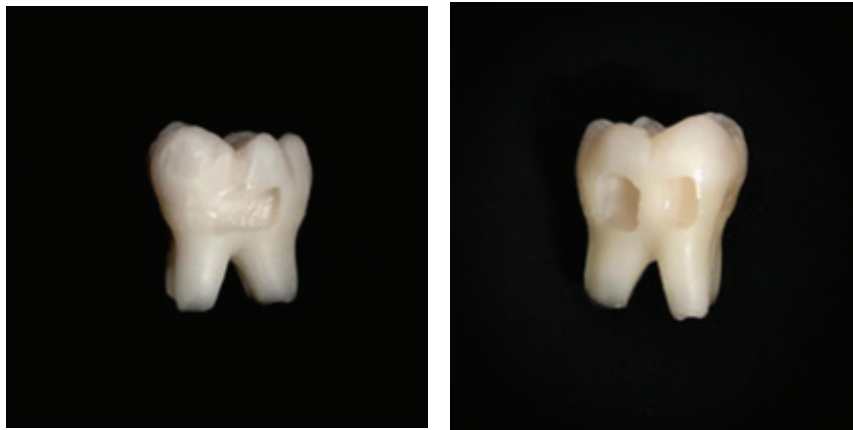


Figure1: A cavity prepared on the vestibular face/ Two cavities prepared in the palatine face

These restorations were immersed in distilled water at 37 °C for 24 hours and next the samples had their apexes sealed with conventional composite resin to prevent the penetration of the dye into the inner part of the tooth. The surfaces of the teeth were dried and covered with two layers of nail polish (figure 2), with an interval of 20 minutes at each application, to ensure the safe insulation of the entire tooth to prevent dye penetration into areas of microcracks or structural defects that have not been observed during selection of teeth. Except for an area of 1.0mm, covered with tape around the adhesive interface of the restoration, evaluated with millimeter probe. In this way, restricting the penetration of the dye to the margins of the cavity.



Figure 2: Sealing with nail polish

2.2 Specimen Infiltration

Then, the samples were immersed in methylene blue solution for 7 days and afterwards were washed under running water for 10 minutes. To determine dye penetration, the specimens were cut with precision metallographic cutter (IsoMet 1000 Precision Sectioning Saw-Buehler-Brazil) (figure 3). The cuts followed the mesio-distal direction from the center of the restoration with a distance of 1 mm.

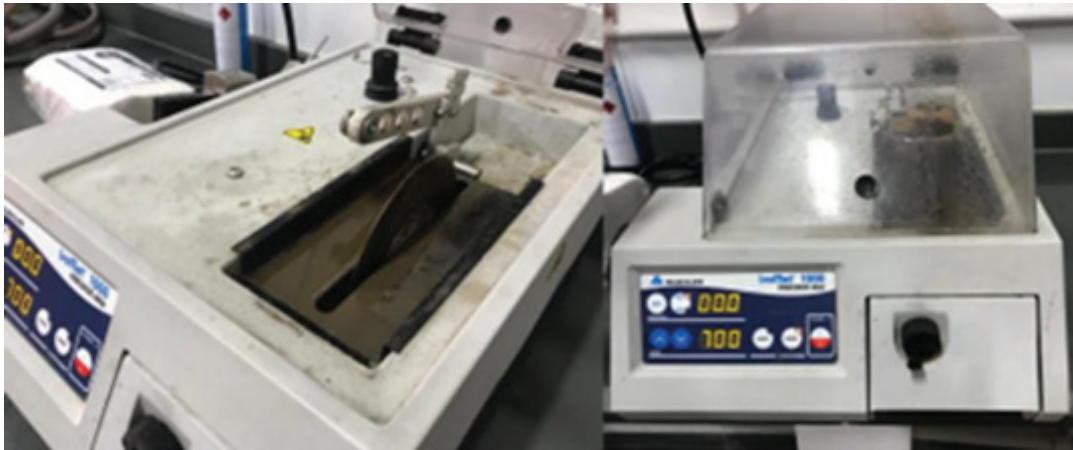


Figure 3: Precision metallographic cutter

3 ANALYSIS OF THE INFILTRATION

Microinfiltration was analyzed according to the following scoring system: 0= without microinfiltration; 1= partial infiltration of the occlusal or cervical wall; 2= total infiltration of the occlusal or cervical wall; 3= total infiltration of the occlusal and axial wall. The factor "ADHESIVE PROTOCOL" (conventional vs. self-conditioning) and the factor "MARGIN IN ENAMEL AND DENTIN" were evaluated. Thus, four reading interfaces were generated, which were evaluated by a calibrated examiner, under an increase of 10x under optical microscope (Carl Zeiss), according to the scores previously established for the determination of the degree of infiltration in the gingival wall of the restorations, with margins in enamel/dentin/cementum.

3.1 Statistical Analysis

The data were submitted to Tukey's non-parametric test, with a significance level of 5%.

3.2 Study blinding

The study was conducted in a double-blind manner, with the treatments being performed by a trained and calibrated operator blinded to the restorative techniques and the evaluation of infiltration was performed by another operator blinded to the groups evaluated.

4 RESULTS

The results obtained through the analysis of the penetration of the dye in enamel and dentin ($p=0.0001$) of the specimens will be presented in the following table (table 1), following the distribution of scores. Group 1 was stipulated, corresponding to selective conditioning (enamel acid), group 2 with total conditioning (enamel acid and dentin) and group 3 without conditioning. When using the Tukey nonparametric test at the significance level of 5%, we observed the sum of the values obtained, presenting the mean between the score data.

Group	Structure	Mean	Homogeneous Data	
Selective conditioning	E	0	B	C
Selective conditioning	D	1,2		
Total conditioning	E	0	B	C
Total conditioning	D	1,6		
Without conditioning	E	2,9	A	
Without conditioning	D	2,9	A	

Table 1: Mean Infiltration/groups

5 DISCUSSION

By demineralizing tooth enamel, acid conditioning provides surface cleaning, increased surface free energy, mineral dissolution and microporosity formation. Later, with the application of the adhesive system, the material infiltration occurs in the previously formed irregularities. In this way, we obtain a durable and resistant micromechanical bond (Swift; Perdigão; Heymann, 1995). This is favored by the absence of moisture from this substrate (Hashimoto *et al.*, 2003). When we refer to dentin, the process of adhering has challenges. Due to the composition of this substrate, we have a lower mineral content, presence of organic matrix and water. This tissue is classified as heterogeneous, having a tubular structure of great regional variation permeable and moist, making the substrate sensitive to the technique used (Chiba *et al.*, 2016; LI *et al.*, 2016; Maravic *et al.*, 2017; Perdigão, 2020). Therefore, there is a significant structural differentiation between the two tissues, which may reflect the differentiated behavior of the adhesive system.

In enamel, the experimental groups that were conditioned (group 1 and group 2) showed no dye at the resin/enamel interface, with infiltration score 0. This fact confirms the statement that the structural characteristics of dental enamel favor micromechanical and clinically stable union when this tissue is conditioned with phosphoric acid (Buonocore, 1955; Swift; Perdigão; Heymann, 1995). The scores obtained for these groups determined a restoration without gaps and with less prevalence of infiltration and side effects.

In relation to dentin, the process of adhering occurs through the formation of the hybrid layer. In this way, it is essential that the mineral phase be removed from the substrate, without altering the collagen matrix, which is later infiltrated by the adhesive material (Anusavice, 2005). For the interface between resin/dentin, in groups 1 and 2, obtained results about marginal infiltration were the same for both techniques. The means obtained were 1.2 and 1.6, respectively, presenting equal statistical analyses.

Despite the similar performance in the face of marginal infiltration, selective conditioning is the technique of choice for dentin adhesion. The acidic monomer of the universal adhesive can remove the mineral and concurrently infiltrate the collagen mesh, preventing part of collagen fibrils that have not been incorporated into the hybrid layer become exposed to degradation (Abad-Coronel; Naranjo; Valdiviezo, 2019).

When comparing the sealing capacity of the groups in enamel and dentin, we obtained significant statistical differences, indicating greater dentin infiltration than in enamel. The literature points out that conditioning with phosphoric acid in dentin hinders the process of adhering. Since the extensive demineralized area generates an insufficient infiltration of resin in the collagen mesh. As a result, there is a decrease in the union resistance, accelerating the penetration of water into the nanometric spaces, characterizing nanoinfiltration (*Chiba et al., 2016; Li et al., 2016; Maravic et al., 2017*).

Another important factor is that in the technique of total acid conditioning the use of phosphoric acid in dentin can cause postoperative sensitivity, insufficient infiltration of the adhesive creating areas of nanoinfiltration, and due to the change in pH that promotes, triggers the activity of metalloproteinases enzymes responsible for the degradation of the hybrid layer (*Abad-Coronel; Naranjo; Valdiviezo, 2019; Perdigão, 2020*). The use of the universal adhesive system by conventional technique affects dentin adhesion because the previous acid conditioning removes mineral and exposes collagen fibers. In this way, the potential chemical adhesion is impaired since functional monomers bind ionically with the calcium of dental tissues (*Swift; Perdigão; Heymann, 1995*). The distribution of scores in group 3 reached values of 2 and 3 in enamel and dentin, characterizing the worst group. The mean obtained by the specimens of group 3 was 2.9, evidencing a statistically significant difference in infiltration, both for dentin and for enamel. The fact that the enamel has not been previously conditioned in this group demonstrates that the demineralizing potential of the self-conditioning adhesive is insufficient, since the mineral dissolution of hydroxyapatite crystals is lower and does not allow the formation of adequate microporosities (*Fujiwara et al., 2018*). In this group dentin infiltration was more expressive, corroborating that if there is no adhesion in resin/enamel, the resin/dentin interface is much more susceptible to degradation (*Vermelho et al., 2017*). Besides the higher prevalence of unfavorable clinical responses in these cases.

According to the results obtained in this study, we can conclude that the integrity of the margins of a restoration is essential to clinical success, since its absence is directly related to the emergence of interface infiltrations. Such infiltrations culminate in several other clinical complications such as: secondary caries, sensitivity and injuries to dentin and pulp.

6 CONCLUSION

The authors conclude that the samples that were not conditioned obtained the highest infiltration rates. Enamel conditioning was better than in dentin and consequently, infiltration was lower in all experimental groups. The selective and total conditioning technique obtained similar results in relation to infiltration.

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