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Original article

Effect of airborne-particle abrasion on dentin with experimental niobophosphate bioactive glass on the microtensile bond strength of resin cements

Edilausson Moreno Carvalho DDS, MSc[a],
Darlon Martins Lima DDS, MSc, PhD[a],
Ceci Nunes Carvalho DDS, MSc, PhD[b],
Alessandro Dourado Loguercio DDS, MSc, PhD[c],
José Roberto Martinelli PhD[d,1], *José Bauer DDS, MSc, PhD* [a,*]

[a] *Department of Dentistry I, School of Dentistry, University Federal of Maranhão (UFMA), Av. dos Portugueses, s/n, 65085680 São Luis, Maranhão, Brazil*
[b] *Department of Restorative Dentistry, School of Dentistry, University Ceuma (UNICEUMA), Josué Montello, 01, Renascença II, 65075120 São Luis, Maranhão, Brazil*
[c] *Department of Restorative Dentistry, School of Dentistry, State University of Ponta Grossa (UEPG), Av. General Carlos Cavalcanti, 4748, Bloco M, Sala 64A, 84030900 Ponta Grossa, Paraná, Brazil*
[d] *Center of Materials Science and Technology, Energy and Nuclear Research Institute (IPEN), Av. Lineu Prestes, 2242, Cidade Universitaria, 05588900 São Paulo, SP, Brazil*

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ABSTRACT

Purpose: The objective of this study was to evaluate the microtensile bond strength (μTBS) of two resin cements bonded to dentin pre-treated with experimental niobophosphate bioactive glass (NBG).

Methods: The experimental bioactive glass was prepared by mixing different amounts of NbO_5; $(NH_4)_2HP_4$; CaO; Na_2CO_3. The particle size distribution and composition of the bioactive glass powder were determined. Twenty flat dentin surfaces from sound extracted human molars were polished with 600-grit SiC paper and air-abraded using experimental bioactive glass niobium powder. The bonding procedures were accomplished by the application of two resin cements: self-etching Panavia F or self-adhesive RelyX U-100. The resin-bonded specimens were cut and the μTBS test was performed after 24 h. The failure mode was determined with a stereomicroscope at 40× magnification. The results were statistically analyzed by two-way ANOVA and Tukey tests ($\alpha = 0.05$).

Results: The two-way ANOVA did not detect interactions between factors, but only a difference between the self-etching and self-adhesive cement ($p = 0.001$). The self-etching resin cement Panavia F obtained a higher μTBS than the self-adhesive cement Relyx U-100. The predominant failure mode of the cements was adhesive/mixed between the resin cement and dentin.

* *Corresponding author at*: Universidade Federal do Maranhão (UFMA), Departamento de Odontologia I, Curso de Odontologia, Rua dos Portugueses, s/n, Campus Universitário do Bacanga, CEP: 65080 805, São Luís, MA, Brazil. Tel.: +55 98 3272 9541; fax: +55 9833018570.
E-mail address: bauer@ufma.br (J. Bauer).

[1] Deceased.

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Conclusion: A new bioactive glass containing niobium did not interfere with the immediate bonding performance of self-etching and self-adhesive cements.
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1. Introduction

Recent studies have pointed out the fact that bond interfaces with dentin deteriorate after relatively short periods of time (*i.e.*, 6 months). This deterioration is mainly caused by hydrolysis of the material, which leaves the collagen fibrils exposed [1,2]. Degradation of collagen fibrils caused by endogenous metalloproteinases (MMP) that increase the permeability within resin–dentin bonded interfaces over time leads to the exposition of the resin–dentin interfaces and can lead to failure of the restoration.

Therefore, the use of a material that has an inhibitory effect on endogenous collagenolytic/gelatinolytic activity [3] is relevant in dentistry to avoid the otherwise inevitable bond degradation that occurs over time [4]. This degradation of the adhesive interface can lead to decreased retention and resistance of the restored tooth's restorative material, since the adhesive interface may compromise resistance to prostheses dental fracture [5].

Some studies have suggested that the use of chlorhexidine (CHX) may be a potential MMP inhibitor [6]. On the other hand, chlorhexidine (CHX) may form crystal-shaped precipitates containing chloride which compromises the bond strength of resin cements [7].

To solve this problem, various studies have pointed toward the direction of using materials such as bioactive glass [8–11]. Bioglass is a material with the ability to interact with dental tissue, inducing the deposition of Ca/P at the bond interface in the presence of body fluids and saliva [12,13]. Thus, the degradation of polymeric materials during its lifetime [14] may be compensated by exposuring to bioactive glass particles, which would induce mineral deposition to remineralize the collagen network. Therefore, it is possible that bioactive glass at the interface could be a material with caries-preventive effects resulting from its special properties of ion release and remineralization.

Silicate glasses known as 45S5 which basic composition is $45SiO_2$, $24.5Na_2O$, 24.5CaO, $6P_2O_5$ (in mol%) have found ample use in applications of biomaterials due to their apatite deposition capacity, which contributes to osseointegration [15]. Previously investigation has been conducted on the effect of airborne-particle abrasion [9] with bioactive glass on dentin before aplication of dental restoration materials such as glass ionomer [16] or dental adhesive materials [17], to create a "*bioactive smear layer*".

Recently, the addition of niobium in the composition of this bioactive glass was proposed to replace silica, obtaining a niobophosphate bioactive glass (NBG). The presence of niobium results in better chemical durability of phosphate glasses, improves their biocompatibility and increases the radiopacity of the material [18–20]. Niobium is a unique metal oxide that manifests properties of crystallinity and self-assembly, functioning as an efficient nucleator of hydroxyapatite. In contrast with silica, Nb would effectively participates in the reaction of osseointegration due to its high biocompatibility with other Ca–P minerals [21].

The aim of this work was to determine the effect of experimental NBG on the microtensile bond strength of self-adhesive and self-etching resin cements to dentin.

Therefore, it was proposed to evaluate the following hypotheses (1) there would be no difference in microtensile bond strength between the two resin cements, and (2) pretreatment of dentin with experimental NBG associated with Panavia F (self-etch) and RelyX U-100 (self-adhesive) cements would affect their microtensile bond strength.

2. Materials and methods

2.1. *Preparation of the experimental niobophosphate bioactive glass*

NBG [18,19] was prepared by melting mixtures of diammonium phosphate (Reagent Grade – Casa Americana, São Paulo, SP, Brazil), niobium oxide (Optical Grade – Companhia Brasileira de Mineração e Metalurgia, Araxá, MG, Brazil), calcium oxide (Reagent Grade – Casa Americana, São Paulo, SP, Brazil) and sodium carbonate (Reagent Grade – Casa Americana, São Paulo, SP, Brazil). The chemical compounds were mixed in a shaker-mixer for 1 h, placed in an alumina crucible, and heated in an electric furnace (Lindberg/Blue M, Watertown, WI, USA).

The heating rate was 10 °C/min up to 500 °C. The material was then kept in air at this temperature for 30 min to eliminate the volatile products. After this, the material was heated to 1400 °C to completely melt the precursors and kept at this temperature for 20 min for homogenization and degassing to eliminate the bubbles. The liquid was poured into a stainless steel mold and cooled at room temperature. The glass was then crushed in a vibrating system with a tungsten ball (Pulverisette, Fristsch, Idar-Oberstein, Germany) for 30 min [12,13]. After grinding, the resultant glass powder was passed through a series of 150 μm–75 μm–53 μm–38 μm–20 μm sieves (Hogentogler & Co., Inc., Columbia, MD, USA). Only the powder that passed through the 20 μm sieve was then used to make the suspensions.

Samples of the powder were analyzed by EDX (EDX-720, Shimadzu, Tokyo, Japan) to verify the final composition and the check for presence of contaminants. The particle size distribution was determined by laser diffraction using a particle size analyzer (CILAS Model 1064, Compagnie Industrielle des Lasers, Orléans, France).

2.2. *Tooth preparation*

Twenty recently extracted sound human third molars were collected after obtaining informed consent from the patients.

Table 1 – Resin luting cements used in this study.

Material	Manufacturer	Composition		Application technique
Panavia F	Kuraray Medical Inc.	ED Primer (Lot A: 00309A) (Lot B: 00183A)	Primer A: HEMA, 10-MDP, 5-NMSA, water, accelerator Primer B: 5-NMSA, accelerator, water, sodium benzene sulphinate	1. Mix (A + B) and apply for 30 s on dentin Gently air-dry
		Paste A (Lot: 00254C) Paste B (Lot: 0031C)	Paste A: 10-MDP, silanated silica, hydrophobic aromatic and aliphatic dimethacrylate, hydrophilic dimethacrylate photoinitiator, dibenzoyl peroxide Paste B: silanated barium glass, sodium fluoride, sodium aromatic sulfinate, dimethacrylate monomer, BPO	1. Dispense equal amounts of pastes A and B and mix pastes for 20 s. 2. Apply mixture on dentin Light-activate (20 s) the margin on each side of the tooth
RelyX U-100	3M/ESPE	Base: glass fiber, methacrylate phosphoric acid esters, dimethacrylates, silanated silica, sodium persulfate (426343) Catalyst: glass fiber, dimethacrylates, silanated silica, p-toluene sodium sulfate, calcium hydroxide (426343)		1. Mix cement (20 s) 2. Apply mixture on dentin 3. Light activate (20 s) the margin on each side of the tooth.

HEMA: 2-hydroxyethyl methacrylate; 10-MDP: 10-methacryloyloxydecyl dihydrogen phosphate; 5-NMSA: N-methacryloyl 5-aminosalicylic acid; BPO: benzoyl peroxide.

The Local University Review Board reviewed and approved this study.

Flat superficial dentin surfaces were created after removal of the occlusal enamel with a low speed diamond saw (Isomet 1000 – Buheler, IL, USA) under water-cooling. After this, the exposed enamel-free dentin surfaces were polished on wet #600-grit SiC paper for 60 s to standardize the smear layer.

Thirty teeth were divided into four groups according to combinations of the main factors *cement resin* at two levels (self-etching and self-adhesive) and *pretreatment of NBG* at two levels (airborne abrasion with experimental NBG – control). Five teeth were used for each experimental group ($n = 5$).

2.3. Crown preparation

Hand-layered build-ups of resin composite 6 mm thick, in the form of blocks ($n = 20$), were made on the flat dentin surfaces of each tooth by the application of 2 mm thick composite layers (LIS, FGM, Joinville, SC, Brazil). Each increment was light-activated with a halogen light appliance (Optilux 501, Kerr) for 40 s at an intensity of 600 mW/cm^2 (Radiometer, Kerr). One side of the composite block was abraded with 600-grit SiC paper under water cooling to create a flat surface with standardized roughness. The composite blocks were ultrasonically cleaned (BioFree 2L, Gnatus Ltda, Ribeirão Preto, SP, Brazil) in distilled water for 10 min and completely air dried. One coat of a silane solution (Silano, Dentsply, Petropolis, RJ, Brazil) was then applied and blow dried before bonding.

2.4. Dentin pretreatment

The airborne-particle system used to deliver the bioactive glass onto the dentin surface was a Jet Star (Microdont, London, UK) unit, used at an air pressure of 0.4 MPa for 15 s at a distance of 1 cm from the dentin surface. The control group did not receive the pretreatment with experimental bioactive glass.

Two resin cements were used: Panavia F (Kuraray Medical Inc., Okayama, Japan) as a representative self-etching type, and RelyX U-100 (3 M/ESPE, St. Paul, MN, USA) as a representative self-adhesive type. The composition and application methods are described in Table 1.

After cementation, a pressure of 5 N was applied to the resin composite blocks with a weight for 5 min [22]. The vestibular, lingual and proximal surfaces were light-activated with a halogen light appliance (Opilux 501, SDS Kerr, USA) for 20 s at an intensity of 600 mW/cm^2 (Radiometer, SDS Kerr, USA). Next, the specimens were stored in distilled water at 37 °C for 24 h.

2.5. Microtensile bond strength (μTBS)

After this, teeth were serially sectioned perpendicular to the adhesive tooth interface into slabs and the slabs into beams with a cross-sectional bonded area of approximately 0.8 mm^2 using a diamond saw (ISOMET 1000 – Buheler, IL, USA). The cross-sectional area of each stick was measured to the nearest 0.01 mm using a digital caliper and recorded (Absolute Digimatic; Mitutoyo, Tokyo, Japan). The number of prematurely debonded sticks (D) per tooth during specimen preparation was recorded but not included in the statistical analysis.

After an interval of storage, the individual bonded sticks were glued onto the Geraldeli device (Odeme, Joaçaba, SC, Brazil) with a cyanoacrylate adhesive (Pegamil Bond Gel, Buenos Aires, Argentina). Each stick was stressed to failure using a universal testing machine (Inston 3342, Cantom, MA, USA) at a crosshead speed of 1.0 mm/min. The maximum tensile load was divided by the specimen's cross-sectional area to express the results in units of stress (MPa).

2.6. Failure pattern analysis

The fractured surface of each test specimen was evaluated under a stereoscopic loupe (Kozo Optical and Electronical

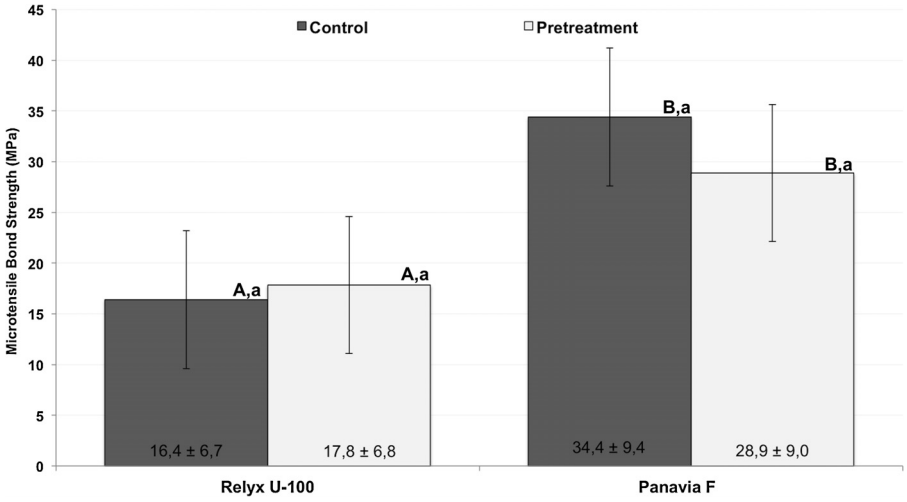

Fig. 1 – The means and standard deviation (MPa) of microtensile bond strength values for all the conditions tested. Data with the same letters (uppercase for cements, lowercase for pretreatment) did not differ significantly ($p < 0.05$).

Instrumental, Nanjing, Jiangsu, China) at 40× magnification and classified as adhesive/mixed (A/M: failure at the resin–dentin interface, which included cohesive failure of the neighboring substrate), cohesive within dentin (CD: failure exclusively within the dentin) or cohesive within materials (CR: failure exclusively within the resin cement or composite). The failure modes were expressed as percentages (%).

2.7. *Statistical analysis*

Statistical analysis was performed using the SigmaPlot 12 software (SigmaPlot v. 12.3, Systat Software Inc., San Jose, USA). Before submitting the data to analysis using the appropriate statistical test, the Kolmogorov–Smirnov and Levene's test performed. After observing the normality of the data distribution ($p = 0.297$) and the equality of the variances ($p = 0.784$), the statistical analysis was perfomed.

The mean μTBS (MPa) of all sticks from the same tooth were averaged for statistical purposes. The μTBS means were analyzed with two-way ANOVA (*Resin cements vs. Pretreatment*) and Tukey tests for pair-wise comparisons ($\alpha = 0.05$).

3. Results

From the EDX data the composition of the niobium phosphate bioactive glass NBG was determined to be 40.1% Nb_2O_5, 32.8% P_2O_5, 21.2% CaO, Al_2O_3 3.8% and 2.1% Na_2O. The NBG particle size distribution showed a large number of particles with diameters less than 20 μm.

The averages and standard deviations of bond strength values are shown in Fig. 1. The two-way ANOVA did not detect statistically significant differences either for the interaction between Cements and NBG Pretreatment ($p = 0.349$) or for the NBG Pretreatment ($p = 0.580$), but only between the self-etching and self-adhesive cement ($p = 0.001$). The self-etching resin cement Panavia F yielded μTBS values statistically superior to those of the self-adhesive cement Relyx U-100.

The failure mode distributions under all conditions tested are depicted in Fig. 2. The predominant failure mode of the cements was adhesive/mixed between the resin cement and dentin.

4. Discussion

Based on the results obtained in this study the resin cements present statistically significant difference between them. Since the self-etching Panavia F resin cement obtained higher values when compared with the self-adhesive Relyx U-100, it can be assumed that deterioration in the bonding efficacy of resin cements might be related to the presence of moisture contamination on the dentin surface [23].

However, the results found in the present study did not demonstrate any interference whatsoever of the bioactive glass particles on the bond strength of the resin cements.

Among the most frequently used glasses, the Bioglass 45S5, which is a bioactive calcium/sodium phosphate-phyllosilicate glass, reacts with body fluids, involving the formation of hydroxyapatite [$Ca_{10}(PO_4)_6(OH)_2$] and the remineralization of dental hard tissues [24].

The bioactivity of the bioglass 45S5 is derived from its reactions with tissue fluids, resulting in the formation of a hydroxycarbonate apatite layer on the glass. When Bioglass 45S5 comes into contact with body fluids, a rapid leaching of Na^+ and congruent dissolution of Ca^{2+}, PO_4^{3-} and Si^{4+} takes place on the glass surface. A polycondensased silica-rich (Si-gel) layer is formed on the bulk glass, which then serves as a template for the formation of a calcium phosphate (Ca/P) layer on its outer surface [25].

This bioactive glass has no Si in its composition, since this element is not part of the osseointegration, as Si has been replaced by niobium. Results with this new bioactive glass in the process of guided bone formation are promising [19]. Thus, a set of minerals (Ca^{2+} and PO_4^{3-}) important for the deposition of hydroxyapatite in which all the raw materials participate in

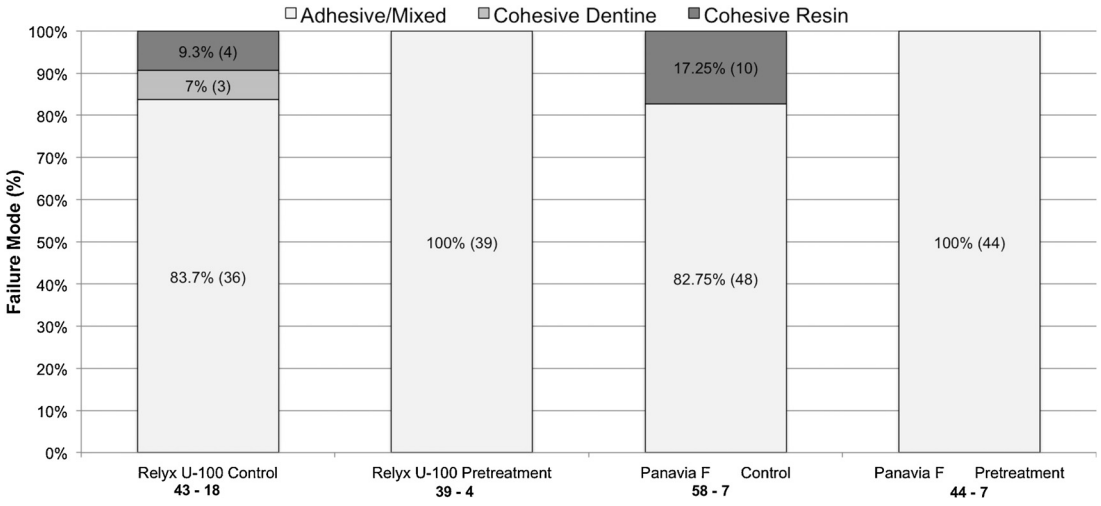

Fig. 2 – Percentagem of failure modes (number of specimens) analyzed by stereoscopic loupe from the control and dentin pre-treated with bioactive glass groups. (Below the columns with the names of the groups: Total specimen number – number of pre-testing failures before the testing.)

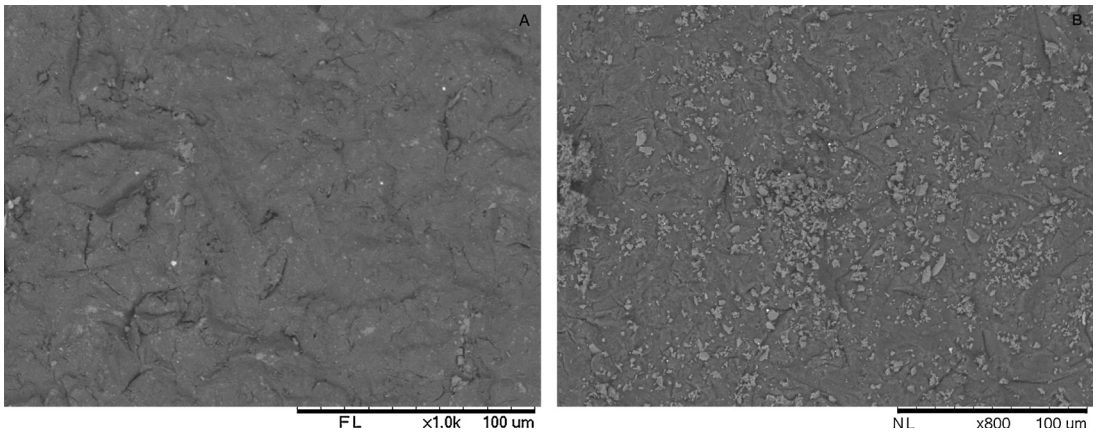

Fig. 3 – (A) Superficial dentin of control group (standard smear layer); (B) formation of "bioactive smear layer" after air-abrasion with NBG on dentin.

the process of osseointegration may be more effective in this new material.

Tamai et al. [26] has shown that bioactive glasses with niobium increased the alkaline phosphatase activity and calcification in osseointegration, with the amount of calcified tissue being directly proportional to the concentration of dissolved niobium ions.

Several authors have used bioglass for the treatment of carious tissue [27,28]. Paolinelis et al. [29] showed that air-abrasion performed with Bioglass 45S5 can be used to prepare both sound and carious dentin, and this technique may be used operatively for the preparation of a noise-, vibration- and pain-free treatment [28]. Other authors have proposed the use of air-abrasion [9,16,17] to produce a "*bioactive smear layer*" formed on the dentin surface during these procedures [30]. Scanning electron microscopy (SEM) pictures shows the difference between control group (Fig. 3A) and the dentin surface treated with an experimental bioactive glass (Fig. 3B).

The present study used only sound dentin, since the focus was on the ability of bioactive glass particles to release minerals (Ca^{2+} and PO_4^{3}), prevent leakage and degradation at the interface and enhance retention of the restoration. The deposition of certain minerals at the bond interface may also contribute to inhibiting the activities of matrix metalloproteinases, resulting in collagenolytic and gelatinolytic activity [10].

Sauro et al. [16] recently showed that air-abrasion pretreatment of dentin surfaces performed with bioglass (45S5, SylcOSspray, London, UK) may induce dentin remineralization and improve the healing ability of restorations performed with light-polymerized resin-modified glass ionomer cements. The group in which dentin was treated only with bioglass showed the lowest microtensile bond strength (immediate) and no statistically significant reduction after 6 months of storage.

In another study, dentin air-abrasion with bioglass did not change the microtensile values after 24 h of a self-etching

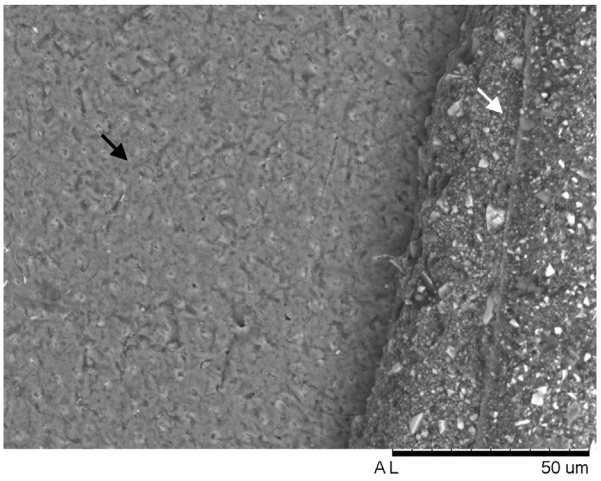

Fig. 4 – Higher magnification of the dentin side of the specimen. The black arrow indicates an area of poor acid conditioning on the dentin and inadequate visualization of dentinal tubules in the Rely X U-100 control group. The white arrow points the layer of resin cement with poor penetration on dentin.

adhesive containing 10-MDP (phosphoric functional monomer), similar in composition to the resin cement Panavia F used in the present study [17].

A similar behavior was shown for the self-adhesive cement Relyx U-100, without any reduction in bond strength values. The preservation of the immediate microtensile bond strength values of the two resin cements may be based on the presence of phosphoric acid modified multifunctional methacrylate monomers (RelyX U-100) and 10-methacryloxydecyl dihydrogen phosphate (Panavia F) that have a high affinity for the minerals in the substrate, in this case, a surface rich in the Ca of NBG.

The self-etching cement (Panavia F) showed the highest bond strength values in the control group, which were close to those found in the literature [31,32]. This cement has chemical bonds and the capacity to remove the smear layer and expose the dentinal tubules, guaranteeing micromechanical bonding. The reason for these high values is because this material contains functional 10-MDP monomer, which has been shown to chemically interact with Ca [33].

In contrast, the self-adhesive Relyx U-100 cement showed the lowest microtensile bond strength. It has been found [34] that this category of luting material interacts only superficially with dental tissues, and the limited flow (due to the cement's viscosity) contributes to reducing infiltration into the dentin tubules (Fig. 4).

Finally, according to the findings of this study, it is proposed that air-abrasion with an experimental bioactive glass is a promising technique to participate in the formation of a ''*hybrid bioactive layer*''. The deposition of a bioactive glass interface could also contribute for various reasons such as: remineralization of the dentin tissue, prevention of collagen degradation by MMP and inhibition of bacterial growth. However, additional studies are needed to confirm these hypotheses.

5. Conclusion

Air-abrasion procedures performed with the use of a new bioactive glass containing niobium did not interfere with the immediate bonding performance of self-etching and self-adhesive resin cements.

Conflict of interest

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

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