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Bond strength of two universal adhesive systems to human dentin using different strategies

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ABSTRACT

The objective of this study was to evaluate the microtensile bond strength (µTBS) to dentin of two universal adhesive systems: Single Bond Universal (SBU) and Ambar Universal (AU), used in different adhesion strategies. ***Materials and Method:*** *Thirty-six human teeth were prepared (n=6) and treated following different adhesive strategies: G1: SBU-etch-and-rinse, applied on dry dentin; G2: SBU-etch-and-rinse, applied on moist dentin; G3: SBU-self-etching; G4: AU-etch-and-rinse, applied on dry dentin; G5: AU-etch-and-rinse, applied on moist dentin; G6: AU-self-etching. The specimens were submitted to µTBS test, failure analysis, and scanning electron microscopy (SEM). Data were analyzed with ANOVA and Tukey's tests (p <0.05).* ***Results:*** *Microtensile bond strength was significantly lower in G1 than G2 and G3. AU adhesive performed worse than the SBU system, except in G5. Cohesive and mixed failures predominated in G1 and G2, while adhesive failures predominated in G3 and G5.* ***Conclusions:*** *Universal adhesives are an interesting innovation, but there are still doubts about their performance, mainly regarding the different protocols provided by the manufacturers. The conventional adhesive strategy on moist dentin demonstrated higher µTBS for both adhesives. The use of the self-etching strategy with the SBU showed promising results.*

Keywords: *dental bonding - dentin - adhesives*

Resistência de união de sistemas adesivos universais à dentina humana usando diferentes estratégias

RESUMO

O objetivo deste estudo foi avaliar a resistência de união à microtração (µTBS) de dois sistemas adesivos universais: Single Bond Universal (SBU) e Ambar Universal (AU), utilizados em diferentes estratégias de adesão. ***Materiais e método:*** *36 dentes humanos foram preparados (n=6) e tratados seguindo diferentes estratégias adesivas: G1: SBU-condicionamento e enxágue, aplicado sobre dentina seca; G2: SBU-condicionamento e enxágue, aplicado sobre dentina úmida; G3: SBU-autocondicionante; G4: AU-condicionamento e enxágue, aplicado em dentina seca; G5: AU-condicionamento e enxágue, aplicado sobre dentina úmida; G6: AU-autocondicionante. Os espécimes foram submetidos ao teste de µTBS, análise de falhas e microscopia eletrônica de varredura (SEM). Os dados foram analisados com os testes ANOVA e Tukey (p<0,05).* ***Resultados:*** *A resistência de união à microtração de G1 foi significativamente menor que G2 e G3. O adesivo AU teve um desempenho pior que o sistema SBU, com exceção do G5. Falhas coesivas e mistas predominaram em G1 e G2 enquanto G3 e G5 apresentaram predominância de falhas adesivas.* ***Conclusões:*** *Os adesivos universais representam uma inovação interessante, mas ainda há dúvidas sobre seu desempenho, principalmente em relação aos diferentes protocolos fornecidos pelos fabricantes. A estratégia adesiva convencional em dentina úmida demonstrou maior µTBS para ambos os adesivos. O uso da estratégia autocondicionante com a SBU apresentou resultados promissores.*

Palabras clave: *adesivos dentinários - dentina - adesivos*

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INTRODUCTION

Adhesion in dentistry is a critical process dependent on numerous factors such as the type of substrate, environment humidity, the adhesive system used, and professional operating capability[1,2]. An adhesive must be able to promote an equally effective bond on both enamel and dentin, even though they are entirely different tissues. On enamel, adhesion occurs due to micro-retention produced on the acid-etched surface filled by the resin monomers. On the heterogeneous dentin substrate, adhesion is challenging because ideal moisture conditions must be maintained to enable adequate infiltration of the adhesive into the demineralized substrate[1-3].

The adhesive systems currently marketed are classified into two categories: 1) conventional (*etch-and-rinse*) and 2) self-etching (*self-etch*)[4]. In conventional systems, the main drawback is the bonding deterioration that occurs in demineralized dentin incompletely filled by the resin monomers after acid etching, leading to microleakage and dentin hypersensitivity that may affect the longevity of a restoration. Self-etching systems were developed to minimize this problem through demineralization and co-occurring primer infiltration[5]. These innovative systems create a chemical interaction between the adhesive and the dental tissue, making the interface more resistant to biodegradation, especially at the dentin substrate[6-7]. Although self-etching adhesive systems do not require moisture to bond, water is included in their composition to ionize the hydrophilic acid monomers, which are responsible for the mineral ions available to the chemical bond with the dental substrate[8-10].

Approximately ten years ago, multi-mode adhesives were designed under the all-in-one concept, providing greater versatility than existing adhesives and enabling the clinician to decide which adhesive strategy to use[1,9,11-14].These universal adhesives tend to minimize dentin sensitivity because deep demineralization is not necessary. Some studies, however, have shown that in enamel, the selective etching technique improves bonding performance[6,8,9,15,16].

Although universal adhesives are an interesting innovation, there are still doubts about their performance, mainly regarding the different protocols provided by the manufacturers[7,17]. Recent systematic reviews concluded that the application of the universal adhesive by the conventional or self-etching method was satisfactory, especially for mild-acidic adhesives[18,19]. However, considering the difficulty of controlling dentin moisture, it is relevant to ascertain whether bond strength changes according to dentin moisture. The aim of this study was thus to evaluate the microtensile bonding strength (µTBS) of two universal adhesive systems applied to human dentin according to different adhesive strategies. This null hypothesis is that different adhesive strategies do not influence bonding strength to human dentin with two adhesive systems.

MATERIALS AND METHOD

Study design and sample number

Two universal adhesive systems, Single Bond Universal (SBU) (3M ESPE, St. Paul, MN, USA) and Ambar Universal (AU) (FGM, Joinville, SC, Brazil) were analyzed with three adhesive strategies: G1: SBU-etch-and-rinse mode and dry dentin; G2: SBU-etch-and-rinse mode and moist dentin; G3: SBU-self-etching; G4: AU-etch-and-rinse mode and dry dentin; G5: AU-etch-and-rinse mode and moist dentin; G6: AU-self-etching.

Considering the error probability of Type I (5%) and Type II (20%), six teeth per group were used, with a minimum of seven specimens per tooth. The teeth prepared were randomly divided into six groups of six teeth, achieving at least 42 specimens (sticks) per group.

Tooth preparation and bonding procedures

This study was approved by the Ethical Committee in Human Research (CEP) (CAAE – 68999817.4.0000.5149). Thirty-six recently removed intact human third molars were selected from the Human Teeth Biobank at the School of Dentistry of the Universidade Federal de Minas Gerais and stored in a 0.5% chloramine solution for 24 hours[20]. The teeth were kept under distilled water until the beginning of the experimental procedures, not exceeding one month after extraction.

A section of the crown was cut perpendicular to the axis of the teeth using a diamond saw blade (Diamond Wafer Blade, Series 15 HC, Lake Bluff, IL, USA) in a cutting machine (IsoMet, Buehler, Lake Bluff, IL, USA) under water cooling, removing the occlusal third of the crown. The dentin surfaces were verified under an optical microscope (Stemi DV4, Zeiss, Oberkochen, Germany) to ensure complete removal of the enamel. To obtain a flat surface of the dental

substrate and create a standard smear layer, all dentin surfaces were polished in a metallographic polisher (Arotec Industry e-Commerce, Cotia, SP, Brazil) with #600 grit silicon carbide abrasive paper (3M, Nova Veneza, SP, Brazil) under water irrigation for 60 s for each tooth before performing the adhesive procedures[21].

The teeth were randomly divided into six groups of six teeth (n = 6), to which were applied the three different adhesive strategies using the two adhesive systems. The adhesives were applied to the flat dentin surfaces according to the manufacturer's instructions (Table 1).

After the adhesive procedure, the dentin surfaces were restored with a composite resin (Filtek Z350 XT, 3M ESPE, St. Paul, MN, USA) to a height of 6 mm, in increments of 2 mm for each layer. Each layer was light-cured for 20 s using a Bluephase (Ivoclar Vivadent, Schaan, Liechtenstein) light-curing device at an intensity of 1.200 mW/cm^2 controlled by a radiometer. At the end of the restorative procedure, the specimens were immersed in distilled water and stored in an incubator at 37 °C for 24 hours.

Specimen preparation

After 24 hours, a diamond saw blade 15.2 cm in diameter and 0.3 mm thick, mounted in the cutting machine, was used under constant water irrigation, with pressure 50 g, and rotational speed 250 rpm, to make sequential cuts in the vestibule/palatal direction, leaving sufficient thickness to obtain slices of approximately 1 mm each considering the

Table 1. Adhesive systems and strategies used

Adhesive System	Composition	Classification according to pH	Groups	Adhesive Strategy
Single Bond Universal (SBU)	10-MDP phosphate monomer, Vitrebond copolymer, HEMA, BISGMA, dimethacrylate resins filler, silane, initiators, ethanol, water	Mild (pH=2.7)	G1	Etch-and-rinse mode and dry dentin: Acid conditioning for 15s; rinse for 30s; air dry for 10s, keeping the dentin dry. Apply the adhesive for 20s with vigorous agitation; gently air for 10s and light-cure for 10s.
			G2	Etch-and-rinse mode and moist dentin: Acid conditioning for 15s; rinse for 30s; remove excess with absorbent paper, keeping dentin moist. Apply the adhesive for 20s with vigorous agitation; gently air for 10s and light-cure for 10s.
			G3	Self-etching: Without acid conditioning. Keep dentin dry, without overdrying it. Apply the adhesive for 20s with vigorous agitation; gently air for 10s and light-cure for 10s.
Ambar Universal (AU)	UDMA, HEMA, methacrylate hydrophilic monomers, methacrylate acid monomers, ethanol, water, silanized silicon dioxide, camphorquinone, ethyl 4-dimethylaminobenzoate, surfactant, sodium fluoride	Mild (pH=2.47)	G4	Etch-and-rinse mode and dry dentin: Acid conditioning for 15s; rinse for 30s; air dry for 10s, keeping the dentin dry. Apply the adhesive for 10s with vigorous agitation, followed by re-application for 10s; gently air for 10s and light-cure for 10s.
			G5	Etch-and-rinse mode and moist dentin: Acid conditioning for 15s; rinse for 30s; remove excess with absorbent paper, keeping dentin moist. Apply the adhesive for 10s with vigorous agitation, followed by re-application for 10s; gently air for 10s and light-cure for 10s.
			G6	Self-etching: Without acid conditioning. Keep dentin dry, without overdrying it. Apply the adhesive for 10s with vigorous agitation, followed by re-application for 10s; gently air for 10s and light-cure for 10s.

HEMA: 2-hydroxyethyl methacrylate; BISGMA: Bisphenol A-glycidyl methacrylate; UDMA: urethane dimethacrylate.

thickness of the disk. Subsequent cuts were made in the mesiodistal direction, maintaining a thickness of 1 mm. After that, cuts were made parallel to the occlusal plane, thereby obtaining stick-shaped specimens with an area of approximately 1.0 mm². The intact specimens (sticks) for each group were measured with a Mitutoyo digital electronic caliper (Kawasaki, Kanagawa, Japan) with a precision of 0.01 mm, confirming the total surface area of approximately 1.0 mm². Sticks with suspected adhesive failure were discarded. The composite resin portion on each stick was identified with a red marker and the dentin portion with a black marker. This procedure facilitated the identification of the parts after fracture. The sticks were stored in distilled water at room temperature until testing.

Microtensile bond strength test

The sticks were individually attached by their ends with a quick-curing cyanoacrylate-based gel adhesive (Super Bonder, Henkel Loctite Adesivos, São Paulo, SP, Brazil) to the Geraldeli's claw. This µTBS device adapts to the specific attachment used in the universal testing machine[22].

The panel of the universal testing machine (EZ -Test, Shimadzu, Japan) was set at a constant speed of 0.5 mm/min and adjusted to detect the maximum load value necessary to fracture the specimen (in kilonewton, kn) and return to the zero (initial) position, after which a new specimen could be positioned for the test. The µTBS results were expressed in MPa and recorded in a spreadsheet.

The number of prematurely detached sticks in each group was recorded, but these values were not included in the statistical analysis. All premature failures that occurred during the cutting procedure and did not exceed 3% of the total number of tested specimens and were similarly distributed among the various groups.

Failure mode analysis and scanning electron microscopy (SEM) analysis

The fractured specimens were observed under the light of a Stemi DV4 optical microscope (Zeiss, Oberkochen, Germany) at 32x magnification by two professionals other than the one who performed the µTBS test. The fracture mode was classified as adhesive (A), mixed (M), cohesive at the resin (CC), or cohesive at dentin (CD). The percentage of failure patterns was calculated according to the frequency observed in each experimental group.

Representative fractured specimens of each group were dehydrated in alcohol in an ascending series (25%, 50%, 75%, 90%, and absolute) for one hour in each solution, followed by immersion in Bis(trimethylsilyl)amine (HMDS) for 10 min. After dehydration, the specimens were fixed on stubs with the aid of a double-sided carbon tape, and sputter-coated with carbon a vacuum sputter-coater (SDC 050, Bal-tec AG, Balzers, Liechtenstein), and observed using a scanning electron microscope (Quanta Fei 200, Hillsboro, OR, USA) operating at an acceleration voltage of 5 kV.

Statistical analysis

For statistical analysis of the data, analysis of variance (ANOVA) ($p<0.05$) was performed to verify differences between the groups. Pairwise comparisons were conducted using Tukey's significant difference test (HSD) ($p<0.05$). GraphPad Prism 7 software (La Jolla, CA, USA) was used for statistical analysis.

RESULTS

Regarding µTBS values, there was a statistically significant difference between G1 (SBU on dry dentin) and G2 (SBU on moist dentin), and between G1 and G3 (SBU in self-etching mode). Table 2 shows the mean and standard deviation of the µTBS test for all groups. SBU applied on moist dentin presented the highest results, followed by the self-etching technique.

Table 2. Means and standard deviation of microtensile bond strength (MPa) of the experimental groups

Groups	Number of specimens	Mean ± SD
G1	71	41.12 (10.72) [a]
G2	77	48.05 (10.27) [b]
G3	61	46.83 (12.87) [b]
G4	8	2.95 (//)
G5	72	44.46 (12.36) [ab]
G6	0	0.00

G1: Single Bond Universal – conventional mode on dry dentin; G2: Single Bond Universal – conventional mode on moist dentin; G3: Single Bond Universal – self-etching mode; G4: Ambar Universal – conventional mode on dry dentin; G5: Ambar Universal – conventional mode on moist dentin; G6: Ambar Universal – self-etching mode
SD: standard deviation; MPa: megapascal; Different letters indicate statistically different means (ANOVA and Tukey's test; $p < 0.05$)

It was impossible to obtain specimens from G6 because the resin became detached from all the teeth during preparation. In G4, only eight specimens were obtained from one tooth, because the resin became detached while the other specimens were being prepared. Hence, G4 and G6 were not included in the statistical analysis. AU applied on moist dentin (G5) showed µTBS similar to SBU.

Figure 1 shows the frequencies of failure modes for each group. Adhesive failure was predominant for SBU in the self-etching mode and for AU in the conventional mode in moist dentin. There were more cohesive and mixed fractures in the SBU specimens when the dentin was etched with phosphoric acid. The SEM images illustrate the predominant failure pattern found in G1, G2, G3 and G5 (Fig. 2)

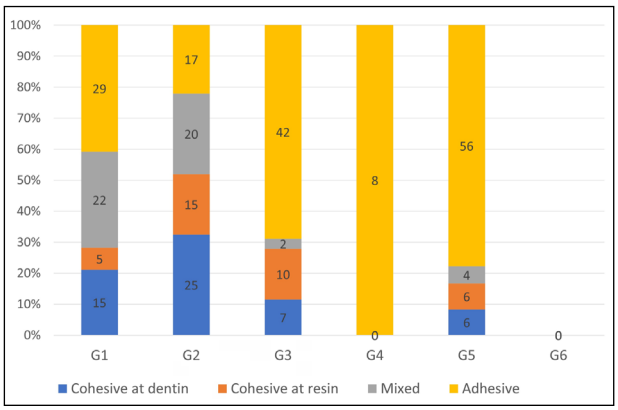

Fig. 1: *Number and percentage of specimens (%) according to the fracture mode of all experimental groups.*
G1= Single Bond Universal - conventional mode on dry dentin; G2= Single Bond Universal-conventional mode on moist dentin; G3= Single Bond Universal-self-etching mode; G4= Ambar Universal-conventional mode on dry dentin; G5= Ambar Universal-conventional mode on moist dentin; G6= Ambar Universal-self-etching mode.

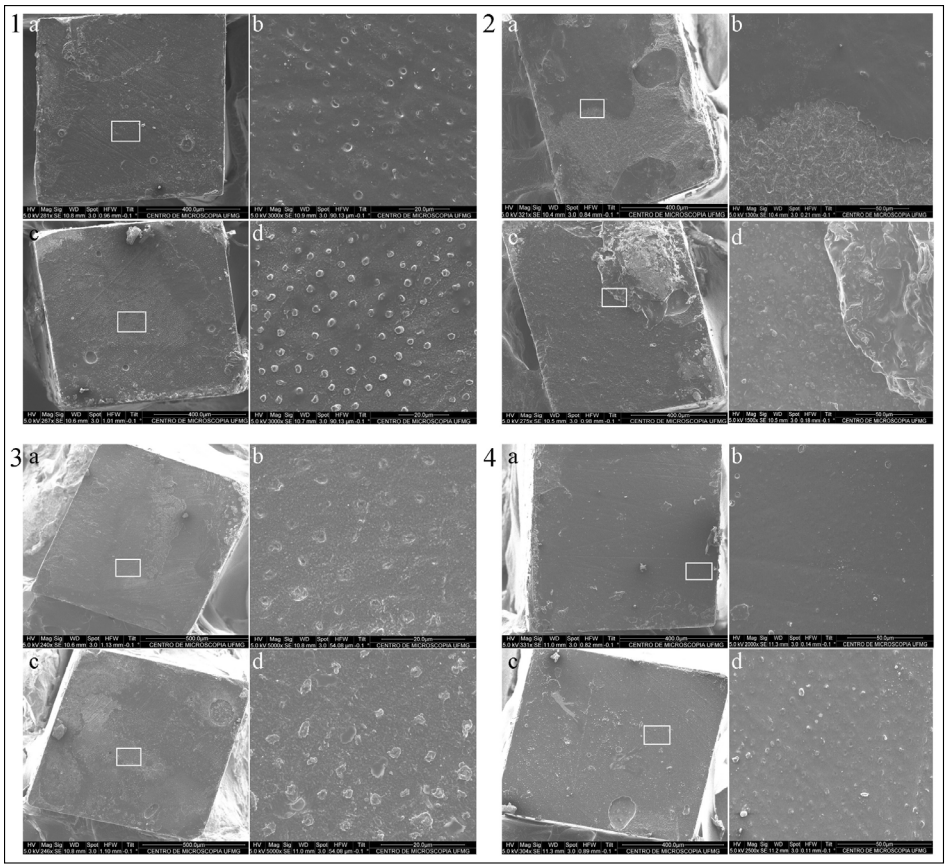

Fig. 2: 1) *SEM images of Single Bond Universal - conventional mode on dry dentin:* ***a-*** *top of the stick, dentin portion, after adhesive-type fracture at 50x and* ***b-****1000x magnifications;* ***c-*** *top of the stick, resin portion at 50x and* ***d-*** *1000x magnifications.* ***2)*** *SEM images of Single Bond Universal - conventional mode on moist dentin:* ***a-*** *top of the stick, dentin portion, after mixed-type fracture at 50x and* ***b-*** *1000x magnifications;* ***c-*** *top of the stick, resin portion at 50x and* ***d-*** *1000x magnifications.* ***3)*** *SEM images of Single Bond Universal - self-etching mode:* ***a-*** *top of the stick, dentin portion, after adhesive-type fracture at 50x and* ***b-*** *1000x magnifications;* ***c-*** *top of the stick, resin portion at 50x and* ***d-*** *1000x magnifications.* ***4)*** *SEM images of Ambar Universal - conventional mode on moist dentin:* ***a-*** *top of the stick, dentin portion, after adhesive-type fracture at 50x and* ***b-*** *1000x magnifications;* ***c-*** *top of the stick, resin portion at 50x and* ***d-*** *1000x magnifications.*

DISCUSSION

This study examined the µTBS of two universal adhesives, Single Bond Universal (SBU) and Ambar Universal (AU). In addition to the self-etching protocol, their behavior was analyzed using the conventional adhesive protocol, with previous conditioning with phosphoric acid (37%) and varying the humidity of the dentin substrate. Although there are systematic reviews of universal adhesive performance, different brands are rarely used, increasing the risk of bias[19]. Thus, we decided to evaluate one system that is used around the world and another Brazilian system that is widely used within the country, mainly due to its low cost, though there are still few studies on it in the literature.

Microtensile tests enable the analysis of the bond between surfaces in small areas[23] using a small number of teeth, considering the possibility of obtaining several replicas, and good customization of the study design. The quantitative analysis of the materials' bond strength to the point of failure can be combined with microscopy techniques to identify the fracture mode at the adhesive interface[24].

During the sectioning of the dentin/resin blocks to produce the specimens (sticks), dentin/resin detachment was observed in the AU self-etch group (G6) and in the AU etch-and-rinse on dry dentin (G4), resulting in zero and eight specimens, respectively. It is worth noting that that research protocols were performed carefully, as described above. Randomization and blocking principles were also followed, ensuring that the sources of variation would be acting comparably in all groups.

We found a statistically significant difference between G1 (SBU on dry dentin) and G2 (SBU on moist dentin), and between G1 and G3 (self-etching SBU). In both comparisons, µTBS results were lower for SBU on dry dentin. These allow us to partially reject the null hypothesis that using different adhesive strategies would not affect µTBS values. However, it is worth highlighting that all results from these adhesives were considered acceptable.

The present study corroborated previous reports that SBU did not differ in self-etching and conventional protocols and had bond strength values similar to ours[25-27]. In contrast, lower µTBS values were found using SBU in the self-etching strategy than in the conventional modes in wet and dry dentin[28]. This can be explained by the higher testing speed applied for the µTBS test (5.0 mm/min) compared to our study and others that used 0.5 to 1.0 mm/min. A clinical trial showed that after 5 years, the clinical behavior of SBU in the etch and rinse strategy was better than in the self-etch strategy, even considering different dentin moisture levels[29]. However, in the current in vitro study, no difference was found between moist and dry dentin.

It should be emphasized that AU did not promote satisfactory bonding using either adhesive strategy, so it was not possible to perform the bond strength test. G5 was the exception, since its results did not differ from those of SBU. Our results contradict other studies that found similar µTBS for the conventional and self-etching protocols with AU[27,30]. Additionally, in these studies, AU did not differ from SBU. However, in contrast to our study, one of them used eroded dentin[30], while another used bovine teeth and considered only adhesive and mixed failures in the calculation of bond strength values[27]. No other study on Ambar Universal was found, but regarding longevity, AU had lower dentinal bond strength after 6 months, and a more stable dentin bond when applied in the etch-and-rinse mode[27].

Adhesives used in self-etching mode are designed to bond to tooth substrates by self-conditioning and simultaneous replacement by resin, integrating the smear layer to the adhesive interface[31]. The ability to infiltrate the smear layer and hybridize the underlying dentin is a process dependent on both the aggressiveness of the self-etching adhesive and the thickness of the smear layer[32].

SBU and AU have similar compositions. They both contain 10-methacryloxydecyl dihydrogen phosphate (MDP) as a functional monomer. Despite sharing similarities in composition and versatility, they can differ in aspects such as the amount of water, solvent, MDP, resin dimethacrylates, and acidity. These differences may influence the viscosity and moisture of the adhesive, affecting its ability to penetrate and act on demineralized or non-demineralized dentin. In the present study, AU did not show positive results when applied in self-etching mode or conventional mode on dry dentin. It was assumed that the acidic monomers were not able to interact sufficiently with the dentin substrate in the self-etching mode to promote adequate demineralization and hybridization. Also, the amount of water may have been insufficient in the conventional mode in dry dentin to promote

rehydration, preventing the adhesive from permeating the collagen network after etching[33].

For SBU, G1 results are compatible with previous reports that consider moisture maintenance essential to achieving successful bonding on conditioned dentin[34-37]. The µTBS proved to be satisfactory, as previous reports allowed us to infer its good capacity to promote rehydration of conditioned and dried dentin[12,28]. SBU contains an ethanol/water-based solvent system with 10–15 wt.% each. Thus, it has enough water to shape the collagen network, promoting re-expansion and re-opening of the interfibrillar spaces from the collapsed dentin, allowing the infiltration of resin monomers[38]. Moreover, the technical profile of SBU indicates that it contains a polyalkenoic acid copolymer (Vitrebond copolymer) capable of providing satisfactory adhesion to the dentin under different humidity levels[39]. The presence of such a substance in AU has not yet been reported. Clinical studies did not show significant differences in the performance of the conventional technique with SBU on dry and moist dentin but found similar marginal adaptation and discoloration for up to 36 months of follow-up[11,15]. Although the performance of SBU in dry dentin in our study was significantly worse, µTBS values were still considerably high. Thus, this may be a valuable option in clinical practice, considering that ideal moisture maintenance in demineralized dentin is challenging to achieve.

In this study, the groups tested with SBU in conventional dry dentin and self-etching modes had prevalence of adhesive- and/or mixed-fracture patterns, with data corroborated by previous reports[28,30]. When acid etching and dry dentin were used, one paper reported predominance of cohesive fracture[12], and another found predominance of cohesive failures for the SBU when tested in both adhesive strategies[27]. In general, studies that tested SBU in conventional mode with moist dentin found a predominance of cohesive failures in dentin or in the composite. These results are reasonable because the higher micromechanical retention obtained after acid etching explains the higher bond strength values, at least in the "immediate" condition. Considering that SBU has not been found to behave differently in the conventional approach in moist dentin and self-etching, its more superficial interaction with the dentin substrate without prior phosphoric acid etching may reduce the risk of postoperative sensitivity and degradation of the collagen fibrils, which could compromise adhesive stability over time[12]. It is believed that this long-term performance is a great advantage of these new adhesive systems.

Although G5 (AU used in conventional mode with moist dentin) had µTBS values that were statistically similar to SBU groups, it is interesting that failures in G5 were predominantly adhesive. Curiously, this behavior is currently observed in the SBU adhesive in self-etching mode. It was expected that failure patterns would be mostly cohesive and/or mixed in the conventional mode, as reported previously[27]. AU bond strength longevity evaluated in bovine teeth showed an increase in the frequency of pre-test failures, especially when used in self-etching mode[27].

There is still no gold standard protocol for universal adhesives[40], which reinforces the importance of continuing to study tools and protocols that improve their use. Long-term clinical trials should be encouraged to confirm the outcomes of universal adhesives.

In conclusion, Single Bond Universal µTBS was higher in etch-and-rinse mode on moist dentin and in self-etching mode compared to etch-and-rinse on dry dentin. Ambar Universal, however, presented acceptable µTBS values only for etch-and-rinse mode on moist dentin.

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DECLARATION OF CONFLICTING INTERESTS

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Isolan CP, Valente LL, Münchow EA, Basso GR, et al. Bond strength of a universal bonding agent and other contemporary dental adhesives applied on enamel, dentin, composite, and porcelain. Applied Adhesion Science. 2014;2(1):1-10. https://doi.org/10.1186/s40563-014-0025-x
2. Pashley DH, Tay FR, Breschi L, Tjäderhane L, et al. State of the art etch-and-rinse adhesives. Dent Mater. 2011;27(1):1-16. https://doi.org/10.1016/j.dental.2010.10.016
3. Pashley DH, Agee KA, Nakajima M, Tay FR, et al. Solvent-induced dimensional changes in EDTA-demineralized dentin matrix. J Biomed Mater Res. 2001;56(2):273-281. https://onlinelibrary.wiley.com/doi/10.1002/1097-4636(200108)56:2%3C273::AID-JBM1095%3E3.0.CO;2-A
4. Tjäderhane L. Dentin bonding: can we make it last? Oper Dent. 2015;40(1):4-18. https://doi.org/10.2341/14-095-BL
5. Miyazaki M, Onose H, Moore BK. Analysis of the dentin-resin interface by use of laser Raman spectroscopy. Dent Mater. 2002;18(8):576-580. https://doi.org/10.1016/S0109-5641(01)00093-8
6. de Goes MF, Shinohara MS, Freitas MS. Performance of a new one-step multi-mode adhesive on etched vs non-etched enamel on bond strength and interfacial morphology. J Adhes Dent. 2014;16(3):243-250. https://doi.org/10.3290/j.jad.a32033
7. Fabião AdM, Fronza BM, André CB, Cavalli V, et al. Microtensile dentin bond strength and interface morphology of different self-etching adhesives and universal adhesives applied in self-etching mode. JAST. 2020;35:723 - 732. https://doi.org/10.1080/01694243.2020.1817722
8. Chen C, Niu LN, Xie H, Zhang ZY, et al. Bonding of universal adhesives to dentine--Old wine in new bottles? J Dent. 2015;43(5):525-536. https://doi.org/10.1016/j.jdent.2015.03.004
9. Luque-Martinez IV, Perdigão J, Muñoz MA, Sezinando A, et al. Effects of solvent evaporation time on immediate adhesive properties of universal adhesives to dentin. Dent Mater. 2014;30(10):1126-1135. https://doi.org/10.1016/j.dental.2014.07.002
10. Perdigão J, Loguercio AD. Universal or Multi-mode Adhesives: Why and How? J Adhes Dent. 2014;16(2):193-194. https://doi.org/10.3290/j.jad.a31871
11. Perdigão J, Kose C, Mena-Serrano AP, De Paula EA, et al. A new universal simplified adhesive: 18-month clinical evaluation. Oper Dent. 2014;39(2):113-127. https://doi.org/10.2341/13-045-C
12. Marchesi G, Frassetto A, Mazzoni A, Apolonio F, et al. Adhesive performance of a multi-mode adhesive system: 1-year in vitro study. J Dent. 2014;42(5):603-612. https://doi.org/10.1016/j.jdent.2013.12.008
13. Lawson NC, Robles A, Fu CC, Lin CP, et al. Two-year clinical trial of a universal adhesive in total-etch and self-etch mode in non-carious cervical lesions. J Dent. 2015;43(10):1229-1234. https://doi.org/10.1016/j.jdent.2015.07.009
14. McLean DE, Meyers EJ, Guillory VL, Vandewalle KS. Enamel Bond Strength of New Universal Adhesive Bonding Agents. Oper Dent. 2015;40(4):410-417. https://doi.org/10.2341/13-287-L
15. Loguercio AD, de Paula EA, Hass V, Luque-Martinez I, et al. A new universal simplified adhesive: 36-Month randomized double-blind clinical trial. J Dent. 2015;43(9):1083-1092. https://doi.org/10.1016/j.jdent.2015.07.005
16. Rosa WL, Piva E, Silva AF. Bond strength of universal adhesives: A systematic review and meta-analysis. J Dent. 2015;43(7):765-776. https://doi.org/10.1016/j.jdent.2015.04.003
17. Miyazaki M, Tsubota K, Takamizawa T, Kurokawa H, et al. Factors affecting the in vitro performance of dentin-bonding systems. Japanese Dental Science Review. 2012;48(1):53-60. https://doi.org/10.1016/j.jdsr.2011.11.002
18. Cuevas-Suárez CE, da Rosa WLO, Lund RG, da Silva AF, et al. Bonding Performance of Universal Adhesives: An Updated Systematic Review and Meta-Analysis. J Adhes Dent. 2019;21(1):7-26. https://doi.org/10.3290/j.jad.a41975
19. Chen H, Feng S, Jin Y, Hou Y, et al. Comparison of bond strength of universal adhesives using different etching modes: A systematic review and meta-analysis. Dent Mater J. 2022;41(1):1-10. https://doi.org/10.4012/dmj.2021-111
20. Muñoz MA, Luque I, Hass V, Reis A, et al. Immediate bonding properties of universal adhesives to dentine. J Dent. 2013;41(5):404-411. https://doi.org/10.1016/j.jdent.2013.03.001
21. Lu S, Zhao SJ, Wang WG, Gao Y, et al. A new fixation method for stick-shaped specimens in microtensile tests: laboratory tests and FEA. J Adhes Dent. 2013;15(6):511-518. https://doi.org/10.3290/j.jad.a29586
22. Perdigão J, Geraldeli S, Carmo AR, Dutra HR. In vivo influence of residual moisture on microtensile bond strengths of one-bottle adhesives. J Esthet Restor Dent. 2002;14(1):31-38. https://doi.org/10.1111/j.1708-8240.2002.tb00145.x
23. Sano H, Shono T, Sonoda H, Takatsu T, et al. Relationship between surface area for adhesion and tensile bond strength--evaluation of a micro-tensile bond test. Dent Mater. 1994;10(4):236-240. https://doi.org/10.1016/0109-5641(94)90067-1
24. Pashley DH, Sano H, Ciucchi B, Yoshiyama M, et al. Adhesion testing of dentin bonding agents: a review. Dent Mater. 1995;11(2):117-125. https://doi.org/10.1016/0109-5641(95)80046-8
25. Muñoz MA, Luque-Martinez I, Malaquias P, Hass V, et al. In vitro longevity of bonding properties of universal adhesives to dentin. Oper Dent. 2015;40(3):282-292. https://doi.org/10.2341/14-055-L
26. Manfroi FB, Marcondes ML, Somacal DC, Borges GA, et al. Bond Strength of a Novel One Bottle Multi-mode Adhesive to Human Dentin After Six Months of Storage. Open Dent J. 2016;10:268-277. https://doi.org/10.2174/1874210601610010268
27. Cardoso GC, Nakanishi L, Isolan CP, Jardim PDS, et al. Bond Stability of Universal Adhesives Applied To Dentin Using Etch-And-Rinse or Self-Etch Strategies. Braz Dent J. 2019;30(5):467-475. https://doi.org/10.1590/0103-6440201902578
28. Leite M, Costa CAS, Duarte RM, Andrade AKM, et al. Bond Strength and Cytotoxicity of a Universal Adhesive According to the Hybridization Strategies to Dentin. Braz Dent J. 2018;29(1):68-75. https://doi.org/10.1590/0103-6440201801698
29. de Paris Matos T, Perdigao J, de Paula E, Coppla F, et al. Five-year clinical evaluation of a universal adhesive: A randomized double-blind trial. Dent Mater. 2020;36(11):1474-1485. https://doi.org/10.1016/j.dental.2020.08.007

30. Siqueira FSF, Cardenas AM, Ocampo JB, Hass V, et al. Bonding Performance of Universal Adhesives to Eroded Dentin. J Adhes Dent. 2018;20(2):121-132. https://doi.org/10.3290/j.jad.a40300
31. Van Meerbeek B, Yoshihara K, Yoshida Y, Mine A, et al. State of the art of self-etch adhesives. Dent Mater. 2011;27(1):17-28. https://doi.org/10.1016/j.dental.2010.10.023
32. Tay FR, Pashley DH. Aggressiveness of contemporary self-etching systems. I: Depth of penetration beyond dentin smear layers. Dent Mater. 2001;17(4):296-308. https://doi.org/10.1016/S0109-5641(00)00087-7
33. Reis A, Loguercio AD, Azevedo CL, de Carvalho RM, et al. Moisture spectrum of demineralized dentin for adhesive systems with different solvent bases. J Adhes Dent. 2003;5(3):183-192. http://www.quintpub.com/userhome/jad/jad_5_3_reis_3.pdf
34. Kanca J, 3rd. Improving bond strength through acid etching of dentin and bonding to wet dentin surfaces. J Am Dent Assoc. 1992;123(9):35-43. https://doi.org/10.14219/jada.archive.1992.0248
35. Tay FR, Gwinnett JA, Wei SH. Micromorphological spectrum from overdrying to overwetting acid-conditioned dentin in water-free acetone-based, single-bottle primer/adhesives. Dent Mater. 1996;12(4):236-244. https://doi.org/10.1016/S0109-5641(96)80029-7
36. Perdigão J, Van Meerbeek B, Lopes MM, Ambrose WW. The effect of a re-wetting agent on dentin bonding. Dent Mater. 1999;15(4):282-295. https://doi.org/10.1016/S0109-5641(99)00049-4
37. Figuerêdo de Siqueira FS, Pinto TF, Carvalho EM, Bauer J, et al. Influence of dentinal moisture on the properties of universal adhesives. International Journal of Adhesion and Adhesives. 2020;101:102633. https://doi.org/10.1016/j.ijadhadh.2020.102633
38. Pashley DH, Tay FR, Carvalho RM, Rueggeberg FA, et al. From dry bonding to water-wet bonding to ethanol-wet bonding. A review of the interactions between dentin matrix and solvated resins using a macromodel of the hybrid layer. Am J Dent. 2007;20(1):7-20. https://www.amjdent.com/Archive/2007/Pashley%20-%20February%202007.pdf
39. 3M. Safety data sheet: 3M ESPE Scotchbond Universal Saint Paul2022 [Available from: https://multimedia.3m.com/mws/mediawebserver?mwsId=SSSSSuUn_zu8l00xMY_eN8mGov70k17zHvu9lxtD7SSSSSS--.
40. Hardan L, Bourgi R, Kharouf N, Mancino D, et al. Bond Strength of Universal Adhesives to Dentin: A Systematic Review and Meta-Analysis. Polymers (Basel). 2021;13(5). https://doi.org/10.3390/polym13050814

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C-shaped Canal System in Maxillary Molars Evaluated by Cone-Beam Computed Tomography in an Argentine subpopulation

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ABSTRACT

The aim of the present study was to evaluate the presence of maxillary first and second molars with a C-shaped canal system in an Argentine subpopulation, and to classify them. ***Materials and Method:*** *Of the 332 CBCTs initially evaluated, 120 met the selection criteria. Once the presence of a C-shaped canal system had been established, the teeth were classified following Martins et al. Data were expressed as absolute frequencies, percentages with of 95% confidence intervals, according to the score test. Comparisons were analyzed by Chi-square test and Fisher' exact test, with 5% significance level.* ***Results:*** *It was found that 5 out of 120 first molars (4%) and 17 out of 152 second molars (11%) had a C-shaped canal system. Regarding the classification applied, of 5 C-shaped first molars, 2 corresponded to type E2 (40%), 2 corresponded to type C (40%), and 1 corresponded to type B1 (20%). Of 17 C-shaped second molars, 4 resembled a type A (24%), 7 resembled a type B1 (41%), 5 resembled a type B2 (29%), and 1 resembled a type C (6%). UC1 and UC2 configurations were the most common at all levels except apical level.* ***Conclusion:*** *The prevalence of C-shaped canal system pattern in maxillary first and second molars was estimated for the first time in an Argentine subpopulation, in vivo. Knowledge of these data should help clinicians during endodontic treatment.*

Keywords: *maxillary molars - C-shaped root canal - cone-beam computed tomography*

Conductos en C en molares superiores evaluados mediante tomografía cone-beam en una sub-población Argentina

RESUMEN

El objetivo del presente estudio fue evaluar la presencia de conductos en C en primeros y segundos molares superiores, en una subpoblación de Argentina. ***Materiales y Método:*** *Se observaron 332 CBCTs, de las cuales 120 cumplieron los criterios de selección. Una vez determinada la presencia de conducto en C se clasificaron según Martins et al. Los datos fueron descriptos mediante frecuencias absolutas y porcentajes, con intervalos de confianza al 95% (IC95), según método score. Las comparaciones fueron analizadas mediante la prueba de Chi-cuadrado o exacta de Fisher con un nivel de significación del 5%.* ***Resultados:*** *De 120 primeros molares, 5 presentaron conductos en C, es decir un 4% y de 152 segundos molares, 17 presentaron conductos en C, es decir un 11%. Según la clasificación aplicada, de 5 primeros molares en C, 2 correspondieron al tipo E2 (40%), 2 al tipo C (40%) y 1 al tipo B1 (20%). De 17 segundos molares en C, 4 pertenecieron al tipo A (24%), 7 al tipo B1 (41%), 5 al tipo B2 (29%) y 1 al tipo C (6%). Las clasificaciones UC1 y UC2 fueron las más representativas en todos los niveles, excluyendo el tercio apical.* ***Conclusión:*** *La prevalencia de conductos en C en primeros y segundos molares superiores, fue estimada por primera vez en una subpoblación Argentina, in vivo. Los datos obtenidos en el presente estudio, resultan de interés para el abordaje de los tratamientos endodónticos.*

Palabras clave: *molares superiores - forma de C - tomografía de haz cónico*

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INTRODUCTION

An understanding of inner dental anatomy and its variations is essential to ensure successful outcomes in endodontic treatment. This is especially significant for the treatment of C-shaped canal configurations, which generally require modifications of the usual instrumentation, irrigation and obturation techniques[1]. When variations of the tooth morphology remain undetected by the clinician, a therapeutic failure can be expected.

The most commonly reported anatomic configuration in maxillary first molars is the presence of 3 roots (2 buccal and 1 palatal) and 4 canals: 2 in the mesiobuccal root, 1 in the distobuccal root and 1 in the palatal root[2]. A maxillary first molar should be considered a four-canal tooth until proved otherwise; however, a clinician should be aware of the possibility of variations in the number of roots, from 1 to 5[3,4]; the number of root canals, from 1 to 8[4-8], or C-shaped root canal configuration which may or may not be split into two or more canals[9]. The first documented case of a maxillary first molar with a C-shaped root canal was published by Newton & McDonald in 1984[10]. According to De Moor[11], the probability of observing a C-shaped canal in the first maxillary molar is 0.091%, although it would be questionable to use the term prevalence in his work, since only the fusion of the distobuccal and palatal roots were considered. In 2006, Cleghorn et al. observed C-shaped canals in 0.12% of maxillary first molars[2].

Maxillary second molars usually have 3 roots: 1 mesiobuccal, 1 distobuccal and 1 palatal, with one canal in each root[12,13], and these roots may be fused[14]. The presence of an extra canal in the mesiobuccal root (MB2) is not as frequent in maxillary second molars as it is in first molars[15]. Different authors have reported variations in the number of roots and canals, and in presence of C-shaped root canals[6,16-18]. Because maxillary and mandibular molars have different anatomical features and numbers of roots, C-shaped root canals in them are classified differently. Martins et al. developed a classification for maxillary molars in 2016[1], since until that date, the definition of a C-shaped maxillary molar configuration was not clear, and no standardization was available.

Cone-beam computed tomography (CBCT) is sufficiently precise for morphological study of the number of roots and canals for diagnostic purposes. It is a useful diagnostic tool in endodontic practice because of its non-destructive *in vivo* application. Additionally, the radiation dose is lower and the resolution is higher than in conventional computed tomography scans, making it more advantageous for clinical application[19].

The aim of this study was to determine the prevalence and the characteristics of C-shaped root canals in maxillary first and second molars, assessed by the CBCT scans taken at the Department of Diagnostic Imaging, School of Dentistry of the University of Buenos Aires (FOUBA), in the Autonomous City of Buenos Aires, Argentina.

MATERIALS AND METHOD

Sample selection

This was a retrospective, observational, cross-sectional, descriptive study which assessed a total 332 CBCT images obtained between August and September 2020. Informed consent was provided by all patients included in this study. The protocol was approved by the Ethics Committee of the School of Dentistry of the University of Buenos Aires (CETICA/FOUBA 006/2020).

Digitalized CBCT images of maxillary first and second molars were collected from a data bank in Diagnostic Imaging Department. During 2020, a total 80,000 patients received care at the FOUBA during the COVID-19 pandemic, and 4,000 CBCT scans were taken, adding up a total of 220,000 services provided.

Inclusion criteria

Maxillary CBCT scans showing at least one molar other than the third molar and developed apices.

Exclusion criteria

Teeth with crown-root decay involving pulp chamber floor, previous endodontic treatment, root resorption, artifacts preventing appropriate visualization in images, including crown post/core and/or crown, or faulty radiographic technique.

Radiographic technique and image assessment

The assessed images were acquired with a Planmeca ProMax® 3D Max CBCT system (Planmeca OY, Helsinki, Finland), with 88 kV and 9.0 mA, exposure time 12.07 seconds and voxel size 150-200

µm. The images were assessed with the software corresponding to the tomograph in shifts of only two hours a day, to prevent visual strain and any misinterpretation of images. The collected data were entered in ad-hoc data recording sheets.

C-shaped root canal classification

This study was based on the classification proposed by Martins et al.[1], which says that a canal is considered as C-shaped in a maxillary molar when it exhibits root fusion and 3 consecutive axial cross sections with an upper-C (UC) 1 or 2 configuration in the fused root. The UC configuration system for maxillary molars is a modification of the Fan et al.[20] classification for C-shaped mandibular molars, and 5 configurations were observed in axial slices: UC1, continuous large C-shaped canal system; UC2, continuous C-shaped canal with 2 main canal lumens in the extremities connected by a large isthmus; UC3, 2 separate root canals; UC4, a single round or oval root canal; UC5, no canal lumen[1].

In turn, depending on which roots were fused, they were classified into 5 types: Type A: Fusion between the mesiobuccal and palatal roots, forming a semilunar mesiopalatal root canal. Type B: Fusion between the mesiobuccal and distobuccal root canals, forming a semilunar buccal root canal system; the concavity of the semilunar shape may be turned to the palatal (subtype B1) or buccal (subtype B2) root. Type C: Fusion between the distobuccal and palatal roots, forming a large semilunar distopalatal root canal. Type D: Presence of a large palatal root canal, forming a semilunar shape; this type has been previously described as a fusion between 2 palatal roots. Type E: Fusion among the 3 roots; this configuration resembles the mandibular C-shaped anatomy, with a large semilunar mesiopalatal canal merging together with an independent distobuccal canal at a single apical foramen (subtype E1) or with a large semilunar distopalatal canal connecting with a mesiobuccal canal at a single apical foramen (subtype E2)[1].

The classification was developed by observing axial slices at five levels: a) coronal-2 mm apical to the canal orifice openings in the chamber floor; b) apical-2 mm above the anatomic apex; c) middle-middle distance between "coronal" and "apical"; d) one third-middle distance between "coronal" and "middle"; e) two thirds-middle distance between "middle" and "apical".

Statistical analysis

CBCT images were examined by two FOUBA endodontists trained in the observation of tomography slices and updated by means of the continuous critical reading of scientific reports related to the subject matter of this work. Cohen's kappa unweighted coefficient was used to measure interobserver agreement. The kappa coefficient (κ) with a 95% confidence interval (CI95) was obtained. A Z-test was applied to analyze the difference between the coefficient obtained and the zero value, with a significance level of 5%: p-value < 0.05 indicates that the Cohen's kappa coefficient differs significantly from zero. The Cohen's kappa coefficient value was computed according to the criteria proposed by Altman[21]. The assessment was done in software R version 4.0.3[22] using the packages *irr*[23] and *psych*[24].

The age of the patients was described by the following measurements: mean, standard deviation (SD), median, first quartile (Q_1), third quartile (Q_3), minimum and maximum. The remaining data were described by absolute frequencies (AF) and percentages. The score method was used to estimate 95% confidence intervals (CI95) for percentages[25]. The Chi-square test or Fisher's exact test were used to evaluate the association between categorical variables, as required. When all the expected frequencies were higher than or equal to 5, the Chi-square test was utilized. If this condition was not fulfilled, the Fisher's exact test was used. A 5% significance level was set up. The following software was used: Calc, de Apache OpenOfficeTM v. 4.1.6[26] and R v. 4.0.3[22].

RESULTS

The results of Cohen's kappa test for reliability revealed significant agreement for the assessment of C-shaped canal configuration ($\kappa = 0.97$; CI95: 0.91 to 1.00; $Z = 13.7$; $p < 0.05$; $N = 200$).

Out of the 332 CBCTs assessed, 120 (36%; CI95: 31% to 41%) were from patients who met the selection criteria. Out of the 120 patients included in the study, 69 were women (58%; CI95: 49% to 66%) and 51 were men (43%; CI95: 34% to 51%). Age ranged from 19 to 79 years, with mean (SD) 40 years (13) and median (Q_1-Q_3) 38 years (32-48).

A total 272 maxillary molars were assessed; 120 were maxillary first molars (44%; CI95: 38% to 50%) and 152 were maxillary second molars (56%;

CI95: 50% to 62%). A significant association was found between tooth group and presence of C-shaped canal configuration (Chi-square=4.44; df=1; p<0.05; Fig. 1). Out of 120 maxillary first molars, 5 had C-shaped canal configurations (4%; CI95: 2% to 9%), whereas 115 did not (96%; CI95: 91% to 98%). Out of 152 maxillary second molars, 17 had C-shaped canal configurations (11%; CI95: 7% to 17%), whereas 135 did not (89%; CI95: 83% to 93%).

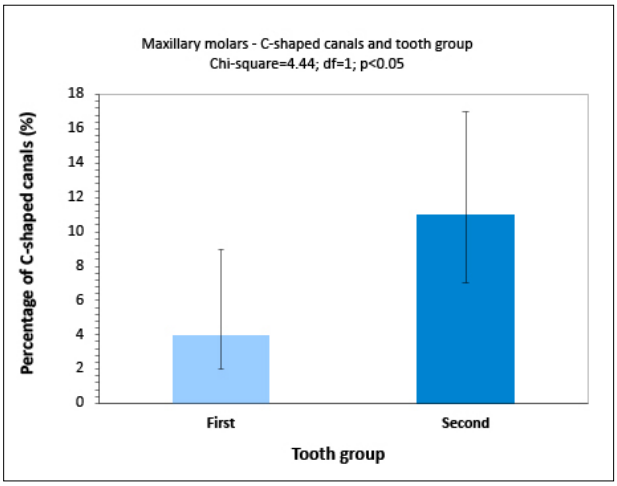

Fig. 1: *Presence of C-shaped canals in maxillary first and second molars (%; CI95).*

The age of patients with C-shaped maxillary first molar canals ranged from 32 to 39 years. The age of patients with C-shaped maxillary second molar canals ranged from 20 to 55 years, with median (Q_1-Q_3) 37 (24-38) and mean (SD) 34 (11) (Fig. 2).

Out of the 5 maxillary first molars with C-shaped canal configuration, 4 belonged to men (80%; CI95: 38% to 96%) and 1 to a woman (20%; CI95: 4% to 62%). Out of the 17 maxillary second molars with C-shaped canal configuration, 7 belonged to men (41%; CI95: 22% to 64%) and 10 to women (59%; CI95: 36% to 78%).

Out of the 22 teeth with C-shaped canals, 11 corresponded to the right side (50%, CI95: 31% to 69%) and 11 to the left side (50%, CI95: 31% to 69%).

In only one of the 2 patients who had both maxillary first molars, a bilateral C-shaped configuration canal was observed, while in the other patient, the C-shaped configuration was seen in tooth 14 (left). In the remaining patients (N=2), the C-shaped configuration was seen unilaterally, 1 in tooth 3 and the other in tooth 14. Regarding the 8 patients who presented both maxillary second molars, 4 had

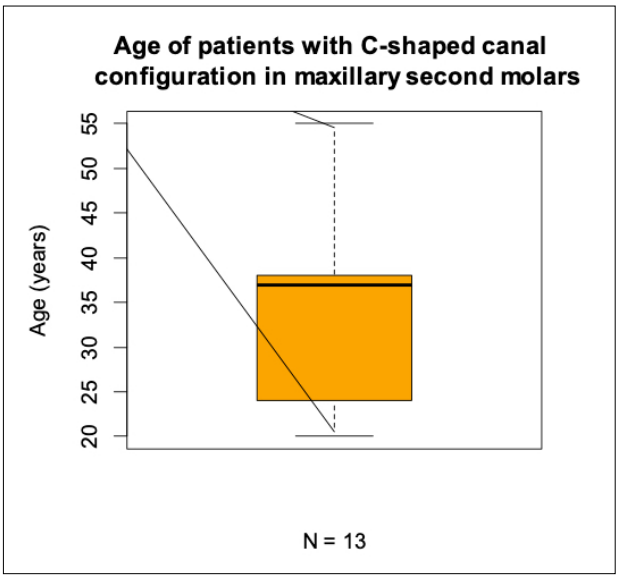

Fig. 2: *Distribution according to age, in patients with C-shaped canal configuration in maxillary second molars. Boxplot: extremes, minimum/ maximum; fences, Q1/Q3; inner line, median.*

bilateral C-shaped configuration canals (N=8 teeth) (50%; CI95: 22% to 78%) (Fig. 3), while the rest did not (50%; CI95: 22% to 78%). In 4 patients out of 9 with unilateral second molars, C-shaped canals were seen in tooth 2 (44%; CI95: 19% to 73%), and in 5 patients, these canals were seen in tooth 15 (56%; CI95: 27% to 81%).

A significant association was found between the C-shaped type according to Martins' classification and the tooth group (Fisher's exact test: p<0.05; Fig. 4). Out of 5 C-shaped maxillary first molars, 2 were type E2 (40%; CI95: 12% to 77%), 2 were type C (40%; CI95: 12% to 77%) and 1 was type B1 (20%; CI95: 4% to 62%). In the single case

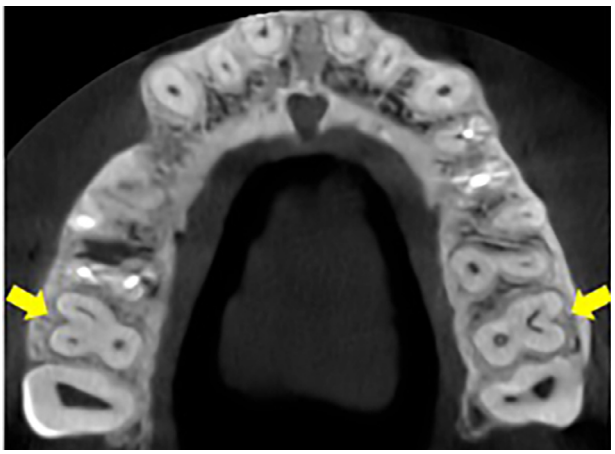

Fig. 3: *Bilateral C-shaped configuration canals in maxillary second molars.*

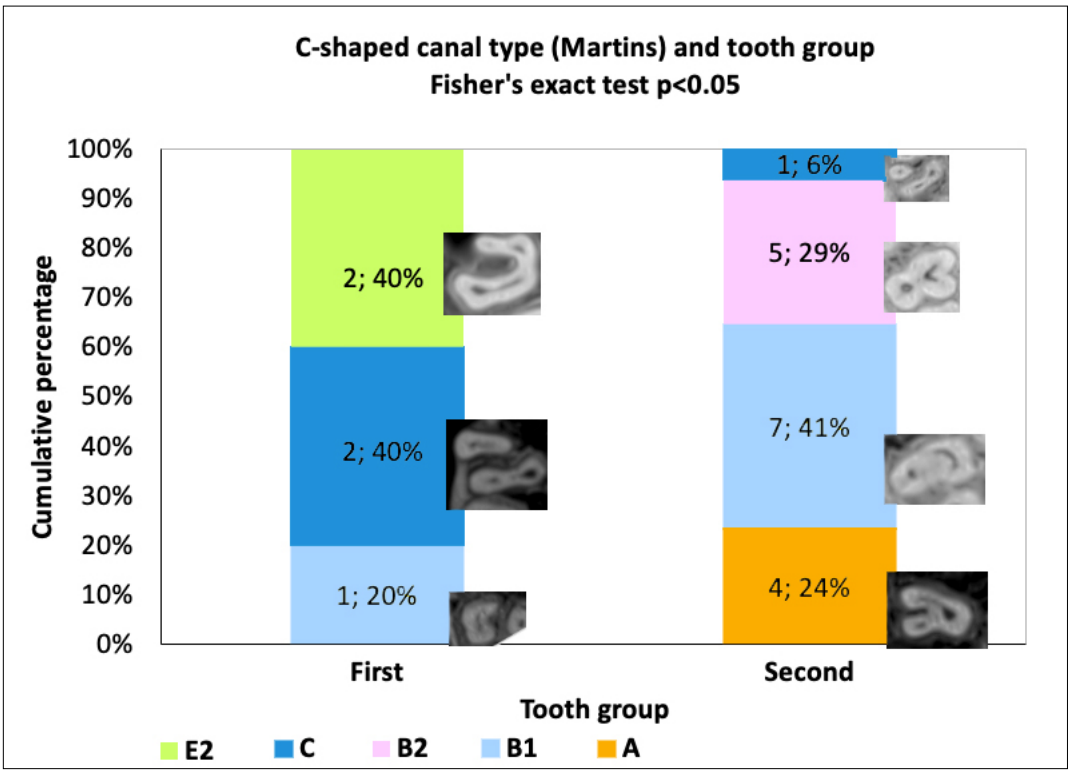

Fig. 4: *Association between the type of C-shaped canal according to the Martins' classification and the tooth group (AF; %).*

of bilateral C, both teeth (3 and 14) had the same configuration. Out of 17 C-shaped maxillary second molars, 4 were type A (24%; CI95: 10% to 47%), 7 were type B1 (41%; CI95: 22% to 64%), 5 were type B2 (29%; CI95: 13% to 53%) and 1 was type C (6%; CI95: 1% to 27%). Out of 4 patients with bilateral C-shaped canals, 2 had the same configuration on both sides, while the other 2 did not.

The UC1 and UC2 categories were the most frequent, except in the apical third, where UC4 was the most frequent. Out of the 5 C-shaped maxillary first molars, 1 was UC1 type (20%; CI95: 4% to 62%) and 4 were UC2 type (80%; CI95: 38% to 96%). Out of 17 C-shaped maxillary second molars, 12 were UC1 type (71%; CI95: 47% to 87%) and 5 were UC2 type (29%; CI95: 13% to 53%). Despite these differences, no statistically significant association was found between the UC configuration and the tooth group (Fisher's exact test: p=0.12). Root fusion was observed in both groups of teeth, but not with the C-shaped configuration.

DISCUSSION

The present study, based on a retrospective assessment of CBCT images, provides a description of the C-shaped anatomy of the first and second maxillary molars, estimated for the first time in an Argentine subpopulation.

The term "C-shaped" in maxillary molars is used to describe root canals with large semilunar canal shape that can represent a whole root canal or a partial fusion of two or more canals[7,10,11,27,28].

In different studies, several methods were used to study root canal morphology, including radiographic technics, clearing technique, spiral computed tomography, sectioning technique and microcomputed tomography, all of which have some limitations. The CBCT scans reveal anatomic details of external and internal anatomy, being an important tool for diagnosis and treatment in endodontic practice[4,7,20].

The prevalence of C-shaped canals in a Portuguese population was 1.1% for first molars and 3.8% for second molars. Martins et al. observed higher prevalence in women[1]. In a Korean population, prevalence was 0.8% in first maxillary molars and 2.7% in second maxillary molars[29]. Mashyakhy et al. reported that the prevalence of C-shaped canals in a Saudi Arabian population was 0.6% in first maxillary molars (only 2 out of 354) and 1.1% in

second maxillary molars (4 out of 372), and the only types found were C and B[18]. A prevalence of 4.9% in second maxillary molars was reported by Yang et al. in a Chinese population after the assessment of 309 extracted teeth by the clearing technique[30]. Ordinola-Zapata et al. evaluated 100 second maxillary molars with fused roots by micro-computed tomography (micro-CT), and found C-shaped configuration in 22 specimens (22%) in a Brazilian population[31]. These differences are likely due to participant ethnicity and age, sample size, study methods and the criteria applied to classify the C-shaped canals.

C-shaped maxillary molars may have low prevalence, but they are of high anatomical complexity because of a large isthmus connecting the root canals that are expected to be separate[1]. Concerning clinical implications, the percentage found in the present study provides heretofore unknown information about the prevalence and characterization of C-shaped canals. This is important since ignoring this morphology might lead to endodontic treatment failure. CBCT scans can enhance the understanding of root canal anatomy, with the potential of improving the outcome of endodontic practice.

Each tooth requiring endodontic treatment should be assessed individually, and the patients should be exposed to the lowest possible amount of radiation to obtain the most useful information for proper diagnoses. When complex, unexpected anatomy is found after access, or when the canals cannot be found, intraoperative CBCT imaging is an excellent tool contributing to correct treatment[32].

A limitation of the current study was that the sample was taken from a single center in the country. FOUBA is a national-level referring center, and although studies about dental anatomy performed by means of CBCT were published[33-35]; further research with a larger number of patients is needed to discuss the outcomes found in this subpopulation.

The percentages found in this study, 4% and 11% of C-shaped canal systems in maxillary first and second molars, respectively, are significant data regarding the prevalence of C-shaped canals in maxillary molars of an Argentine subpopulation.

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DECLARATION OF CONFLICTING INTEREST

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Martins JN, Mata A, Marques D, Anderson C, Caramês J. Prevalence and Characteristics of the Maxillary C-shaped Molar. J Endod. 2016 Mar;42(3):383-9. https://doi.org/10.1016/j.joen.2015.12.013
2. Cleghorn BM, Christie WH, Dong CC. Root and root canal morphology of the human permanent maxillary first molar: a literature review. J Endod. 2006 Sep;32(9):813-21. https://doi.org/10.1016/j.joen.2006.04.014
3. Barbizam JV, Ribeiro RG, Tanomaru Filho M. Unusual anatomy of permanent maxillary molars. J Endod. 2004 Sep;30(9):668-71. https://doi.org/10.1097/01.DON.0000121618.45515.5A
4. Gopikrishna V, Bhargavi N, Kandaswamy D. Endodontic management of a maxillary first molar with a single root and a single canal diagnosed with the aid of spiral CT: a case report. J Endod. 2006 Jul;32(7):687-91. https://doi.org/10.1016/j.joen.2005.10.057
5. Albuquerque DV, Kottoor J, Dham S, Velmurugan N, Abarajithan M, Sudha R. Endodontic management of maxillary permanent first molar with 6 root canals: 3 case reports. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2010 Oct;110(4):e79-83. https://doi.org/10.1016/j.tripleo.2010.04.017
6. Kottoor J, Velmurugan N, Sudha R, Hemamalathi S. Maxillary first molar with seven root canals diagnosed with cone-beam computed tomography scanning: a case report. J Endod. 2010 May;36(5):915-21. https://doi.org/10.1016/j.joen.2009.12.015
7. Kottoor J, Velmurugan N, Surendran S. Endodontic management of a maxillary first molar with eight root canal systems evaluated using cone-beam computed tomography scanning: a case report. J Endod. 2011 May;37(5):715-9. https://doi.org/10.1016/j.joen.2011.01.008
8. Martins JNR, Anderson C. Endodontic treatment of the maxillary first molar with five root canals: three case reports. Rev Port Estomatol Med Dent Cir Maxilo Fac 2013;54(1):37–42. https://doi.org/10.1016/j.rpemd.2012.11.004
9. Kharouf N, Haïkel Y, Mancino D. Unusual Maxillary First Molars with C-Shaped Morphology on the Same Patient: Variation in Root Canal Anatomy. Case Rep Dent. 2019 Oct 22;2019:1857289. https://doi.org/10.1155/2019/1857289
10. Newton CW, McDonald S. A C-shaped canal configuration in a maxillary first molar. J Endod. 1984 Aug;10(8):397-9. https://doi.org/10.1016/S0099-2399(84)80162-4
11. De Moor RJ. C-shaped root canal configuration in maxillary first molars. Int Endod J. 2002 Feb;35(2):200-8. https://doi.

org/10.1046/j.1365-2591.2002.00461.x
12. al Shalabi RM, Omer OE, Glennon J, Jennings M, Claffey NM. Root canal anatomy of maxillary first and second permanent molars. Int Endod J. 2000 Sep;33(5):405-14. https://doi.org/10.1046/j.1365-2591.2000.00221.x
13. Neelakantan P, Subbarao C, Ahuja R, Subbarao CV, Gutmann JL. Cone-beam computed tomography study of root and canal morphology of maxillary first and second molars in an Indian population. J Endod. 2010 Oct;36(10):1622-7. https://doi.org/10.1016/j.joen.2010.07.006
14. Martins JN, Mata A, Marques D, Caramês J. Prevalence of Root Fusions and Main Root Canal Merging in Human Upper and Lower Molars: A Cone-beam Computed Tomography In Vivo Study. J Endod. 2016 Jun;42(6):900-8. https://doi.org/10.1016/j.joen.2016.03.005
15. Silva EJ, Nejaim Y, Silva AI, Haiter-Neto F, Zaia AA, Cohenca N. Evaluation of root canal configuration of maxillary molars in a Brazilian population using cone-beam computed tomographic imaging: an in vivo study. J Endod. 2014 Feb;40(2):173-6. https://doi.org/10.1016/j.joen.2013.10.002
16. Ulusoy OI, Görgül G. Endodontic treatment of a maxillary second molar with 2 palatal roots: a case report. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2007 Oct;104(4):e95-7. https://doi.org/10.1016/j.tripleo.2007.05.027
17. Ozcan E, Aktan AM, Ari H. A case report: Unusual anatomy of maxillary second molar with 3 mesiobuccal canals. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2009 Jan;107(1):e43-6. https://doi.org/10.1016/j.tripleo.2008.09.006
18. Mashyakhy M, Chourasia HR, Jabali A, Almutairi A, Gambarini G. Analysis of Fused Rooted Maxillary First and Second Molars with Merged and C-shaped Canal Configurations: Prevalence, Characteristics, and Correlations in a Saudi Arabian Population. J Endod. 2019 Oct;45(10):1209-1218. https://doi.org/10.1016/j.joen.2019.06.009
19. Patel S, Brown J, Pimentel T, Kelly RD, Abella F, Durack C. Cone beam computed tomography in Endodontics - a review of the literature. Int Endod J 2019;52:1138-52. https://doi.org/10.1111/iej.13115
20. Fan B, Cheung GS, Fan M, Gutmann JL, Bian Z. C-shaped canal system in mandibular second molars: Part I--Anatomical features. J Endod. 2004 Dec;30(12):899-903. https://doi.org/10.1097/01.don.0000136207.12204.e4
21. Altman DG. Practical statistics for medical research. London, UK, Chapman & Hall, 1991. https://doi.org/10.1201/9780429258589
22. R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL: http://www.r-project.org/index.html
23. Gamer M, Lemon J, Singh IFP. irr: Various Coefficients of Interrater Reliability and Agreement. 2019. https://CRAN.R-project.org/package=irr.
24. Revelle W. psych: Procedures for Personality and Psychological Research, Northwestern University, Evanston, Illinois, USA. 2020. https://CRAN.R-project.org/package=psych
25. Newcombe RG, Merino Soto C. Intervalos de confianza para las estimaciones de proporciones y las diferencias entre ellas. Interdisciplinaria. 2006;23(2):141-54. URL: http://www.scielo.org.ar/scielo.php?script=sci_arttext&pid=S1668-70272006000200001&lng=es&tlng=es
26. Apache Software Foundation. Apache OpenOfficeTM v. 4.1.6. 2018. URL: https://www.openoffice.org
27. Dankner E, Friedman S, Stabholz A. Bilateral C shape configuration in maxillary first molars. J Endod. 1990 Dec;16(12):601-3. https://www.jendodon.com/article/S0099-2399(07)80204-4/pdf
28. Martins JN, Quaresma S, Quaresma MC, Frisbie-Teel J. C-shaped maxillary permanent first molar: a case report and literature review. J Endod. 2013 Dec;39(12):1649-53. https://doi.org/10.1016/j.joen.2013.06.032
29. Jo HH, Min JB, Hwang HK. Analysis of C-shaped root canal configuration in maxillary molars in a Korean population using cone-beam computed tomography. Restor Dent Endod. 2016 Feb;41(1):55-62. https://rde.ac/DOIx.php?id=10.5395/rde.2016.41.1.55
30. Yang ZP, Yang SF, Lee G. The root and root canal anatomy of maxillary molars in a Chinese population. Endod Dent Traumatol. 1988 Oct;4(5):215-8. https://doi.org/10.1111/j.1600-9657.1988.tb00324.x
31. Ordinola-Zapata R, Martins JNR, Bramante CM, Villas-Boas MH, Duarte MH, Versiani MA. Morphological evaluation of maxillary second molars with fused roots: a micro-CT study. Int Endod J. 2017 Dec;50(12):1192-1200. https://doi.org/10.1111/iej.12752
32. Ball RL, Barbizam JV, Cohenca N. Intraoperative endodontic applications of cone-beam computed tomography. J Endod. 2013 Apr;39(4):548-57. https://doi.org/10.1016/j.joen.2012.11.038
33. Chaintiou Piorno R, Consoli Lizzi EP, Saiegh J, Vázquez DJ, Gualtieri AF, Rodríguez PA. C-Shaped Canal System in Mandibular Second Molars Evaluated by Cone-Beam Computed Tomography in an Argentine Subpopulation. vGlobal Journal of Medical Research: J Dentistry & Otolaryngology 2019;19:16-23.
34. Chaintiou Piorno R, Consoli Lizzi EP, Gualtieri AF, Rodríguez PA. C-Shaped canal system in mandibular premolars evaluated by cone-beam computed tomography in a population of the Autonomous City of Buenos Aires, Argentina. Arch Oral Biol. 2021 Mar;123:105040. https://doi.org/10.1016/j.archoralbio.2021.105040
35. Consoli Lizzi EP, Chaintiou Piorno R, Aranda CM, Gualtieri AF, Rodríguez PA. Maxillary incisor internal root anatomy evaluated by cone-beam computed tomography in a population of the Autonomous City of Buenos Aires, Argentina. Acta Odontol Latinoam. 2021 Aug 1;34(2):188-194. https://doi.org/10.54589/aol.34/2/188

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Salivary *Streptococcus mutans* colony-forming unit count in patients with and without orthodontic appliances

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ABSTRACT

Orthodontic appliances promote the accumulation of biofilm in the oral cavity and increase counts of Streptococcus mutans (S. mutans). However, there are few comparative studies of the effects generated by the interaction of saliva and microorganisms in absence and presence of orthodontic appliances. ***Aim:*** *The aim of this study was to determine the S. mutans colony-forming unit count (CFU/mL) in participants with and without fixed orthodontic appliances.* ***Materials and Method:*** *It was an observational cross-sectional study on 21 participants, all over 18 years of age, non-smokers, without removable oral appliances, who had not been under antibiotic treatment within the previous three months. Sociodemographic variables, oral hygiene habits, S. mutans CFU/mL count, and salivary pH were assessed. Saliva samples were collected, and the data was analyzed using Fisher's exact and Kruskal Wallis tests. A p-value <0.05 was considered statistically significant.* ***Results:*** *Fourteen (66.7%) of the participants were female; average age was 20.4 ± 2.2 years. The group without fixed orthodontic appliances had the highest salivary S. mutans CFU/mL count (Me: 56.0×103, IQR: 9.2×103 - 75.5×103), but there was no statistically significant difference between groups (p=0.7459). There was a statistically significant difference in salivary pH, with the metal orthodontic appliance group having the lowest pH (p=0.0478). No statistically significant difference in salivary S. mutans CFU/mL count was found between groups. Salivary pH was lower in the group with metal appliances than in the groups with non-metal appliances and without appliances.*

Keywords: *streptococcus mutans - saliva - orthodontics - mouth*

Recuento de unidades formadoras de colonias de *Streptococcus mutans* salivares en pacientes con y sin aparatología ortodóntica

RESUMEN

Se ha reportado que la aparatología ortodóntica promueve la acumulación de biofilm en la cavidad bucal y aumenta los recuentos bacterianos de Streptococcus mutans (S. mutans). Sin embargo, los estudios comparativos sobre los efectos generados por la interacción de la saliva y los microorganismos en ausencia y presencia de aparatología ortodóntica son limitados. Determinar el recuento de Unidades Formadoras de Colonias (UFC/mL) de S. mutans en participantes con y sin aparatología ortodóntica fija. ***Materiales y Método:*** *se realizó un estudio observacional de corte transversal con 21 participantes, todos mayores de 18 años, no fumadores, sin ningún tipo de aparatología oral removible y sin antecedentes de tratamiento antibiótico en los tres meses previos. Se evaluaron variables sociodemográficas, hábitos de higiene oral, recuento de UFC/mL de S. mutans y pH salival. Se recolectaron muestras salivales y los datos se analizaron mediante las pruebas Exacto de Fisher y Kruskal Wallis. Un valor de p ≤0,05 se consideró estadísticamente significativo.* ***Resultados:*** *participaron catorce (66,7%) mujeres; la edad promedio fue de 20.4 ± 2.2 años. El grupo sin ortodoncia fija presentó el mayor recuento de UFC/mL de S. mutans salival (Me: 56,0×103, RIC: 9,2×103 - 75,5×103), pero no hubo una diferencia estadísticamente significativa entre los grupos (p=0,7459). Con relación al pH salival, se observó una diferencia estadísticamente significativa, siendo el grupo de ortodoncia metálica el que presentó el pH más bajo (p=0,0478). Aunque no se encontró una diferencia estadísticamente significativa en el recuento de UFC/mL de S. mutans salival entre los grupos, el pH salival del grupo de aparatología metálica fue más bajo en comparación con los grupos no metálicos y sin aparatología.*

Palabras clave: *streptococcus mutans - saliva - ortodoncia - biopelícula*

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INTRODUCTION

Fixed orthodontic appliances are one of the most common ways to treat dental malocclusions. Given their retentive nature, these appliances have been associated to increased dental biofilm in the oral cavity[1-4]. Dental biofilm is made up of multiple species of acidogenic bacteria related to dental caries progression such as *Lactobacillus spp*. and *Streptococcus mutans (S. mutans)*. These bacteria produce acids as a byproduct of fermentable carbohydrate metabolism, which in a favorable oral environment can lead to demineralization of the tooth surface[5]. The adherence and colonization of these microorganisms during orthodontic treatment is influenced by various factors such as bracket material, its physical properties, the presence and type of ligatures, and the patient's oral hygiene habits[3]. Microbial interactions between the surface of the brackets and the oral microbiota are due in part to the amount of surface free energy (SFE) in the bracket material, which is a determining factor in the degree of bacterial adhesion and therefore also in the accumulation of dental biofilm[6]. Dental literature suggests a positive correlation between adherence of *S. mutans* and orthodontic materials with high SFE[7]. However, the findings are contradictory: Eliades et al. reported higher SFE in metal brackets compared to ceramic brackets, but Lee et al. found the opposite[7,8].

Normally, the oral microbiome is in balance, but the presence of fixed orthodontic appliances can cause a pathological imbalance that leads to a demineralizing process in tooth enamel[5]. A meta-analysis by Sundararaj et al. found that the incidence of white spot lesions in patients with orthodontic treatment was 45.8%, which suggests that fixed orthodontic appliances are associated with increased presence of *S. mutans* and an uncontrolled decrease in salivary and oral pH, which are necessary etiological factors for the initial development of carious lesions[5,9].

Some of the most important factors to evaluate when planning an orthodontic treatment include individual patient risk assessment based on their oral hygiene habits, microbiological and salivary conditions, dental caries experience and the presence of periodontal disease, among others. According to Bonetti et al. (2013), salivary properties are not affected by the presence of fixed orthodontic appliances[10]; nevertheless, Arab et al. (2016) concluded that there are salivary and microbiological changes associated with the presence of fixed orthodontic appliances[11].

In an in vitro study, Brusca et al. observed that *S. mutans* adherence was greater on non-metal brackets (composite and ceramic) than on metal brackets[12]. However, in a similar study, Pappaionnau et al. did not find any difference in bacterial adherence related to bracket material, and therefore recommended conducting an in vivo study to determine whether there is a clinically important difference among various types of brackets, and their effect on the presence of *S. mutans*[13].

Although some studies have been published on the subject, there is still no conclusive evidence about the relationship between the number of salivary *S. mutans* colony-forming units (CFU) in patients undergoing fixed orthodontic treatment and the different types of bracket material, compared to patients without orthodontic appliances. Thus, the aim of this study was to determine the salivary *S. mutans* CFU/mL count in participants with and without fixed orthodontic appliances.

MATERIALS AND METHOD

This was a cross-sectional study on 21 participants. Previous publications were used as a reference for sample size[10,11]. Applying a non-probability convenience sampling method, individuals were invited to participate voluntarily, and those selected for the study were all of legal age, non-smokers, without orthodontic appliances or with metal and non-metal fixed orthodontic appliances. Subjects who had been under antibiotic treatment within the three months prior to sampling or used any type of dental prosthesis or removable oral appliances were excluded.

The following variables were analyzed: sociodemographic (age, sex, socioeconomic status), orthodontic treatment (absence or presence of appliances, metal or non-metal fixed appliances), oral hygiene (frequency of tooth brushing and use of dental floss and mouthwash), microbiological (*S. mutans* CFU/mL count), and salivary pH.

Saliva collection

All samples were collected during the morning to avoid possible alterations due to diurnal variations in salivary pH[14]. The participants were instructed to brush their teeth two hours or more before sample collection and not to eat anything after that. Each

participant was given a piece of bite rim wax (1 cm^3) to chew for three minutes, and two sterile test tubes and funnels. Sufficient time was allotted for the collection of 5 mL of saliva per tube, one for microbiological culture and the other for salivary pH analysis.

Salivary pH analysis

Salivary pH was measured no more than 30 minutes after sample collection using a benchtop pH-meter (Hanna® Instruments HI-2210) that was properly calibrated between each sample. The tip of the electrode was disinfected with fifth-generation quaternary ammonium solution (Benzaldina®). The pH-meter electrode was immersed in the test tube carefully, without touching the bottom, leaving it suspended until the digital measurement was recorded.

Microbiological analysis

The saliva samples were processed in a sterile environment inside a previously decontaminated laminar flow cabinet. Using a calibrated micropipette, 0.5 mL of each sample were aseptically transferred to test tubes with sterile saline solution (4.5 mL), sequentially, to obtain serial dilutions from 10^{-1} to 10^{-3} (dilution factors were determined based on results collected in a pilot study). After every transfer, each suspension was vortexed for sixty seconds.

The samples were cultured in Petri dishes with Mitis Salivarius Agar (Becton, Dickinson and Company, Difco™) supplemented with 0.2 U/mL of bacitracin and potassium tellurite for selective isolation of *S. mutans*[15]. The culture media were checked to ensure absence of condensation. A micropipette was used to transfer 100 µl of each dilution, performing duplicate plating and changing the tip between each dilution. Each drop was spread evenly over the agar surface using a sterile Drigalski spatula, sequentially, from the highest to the lowest dilution factor. Seven plates were cultured for each saliva sample: one direct plating and three dilutions in duplicate.

The petri dishes were incubated in anaerobic jars (ThermoScientific™) with anaerobic gas generating sachets (ThermoScientific™ Oxoid AnaeroGen) for 48 hours at 37 ºC. *S. mutans* CFU/mL were counted using a digital colony counter, identifying each colony based on macroscopic characteristics such as color and morphology. In cases of doubt, Gram staining was used.

The information obtained was entered in duplicate in two excel databases and validated using the Epidata 3.1 software. Any discrepancies were corrected, and a fully refined database was exported to the Stata I/C version 14.0 statistical package for analysis.

Statistical analysis

The univariate analysis of the qualitative variables consisted of calculating absolute frequencies and percentages. For quantitative variables, measures of central tendency and dispersion were calculated. In the bivariate analysis, Fisher's exact test was applied for qualitative variables, and Kruskal Wallis test was used for quantitative variables, as required, considering that the salivary pH and CFU count variables did not have normal distribution. Dunn's test was used as a post-hoc test for multiple comparisons. Statistical significance was considered for p-values lower than 0.05 ($p<0.05$).

Ethical considerations

This study was approved with the Ethical Concept 05222014 by the Ethics in Research Committee of the School of Dentistry at Universidad Santo Tomás (Colombia). It was classified as "research with minimum risk" according to Resolution 8430 of October 1993, which establishes the scientific, technical and administrative standards for health research in Colombia[16]. All participants signed an informed consent form after receiving an explanation of the aim and procedure of the study. The principles of respect for autonomy, beneficence, justice and nonmaleficence were applied.

RESULTS

Of the 21 participants, 14 (66.7%) were females, and all were university students. Mean age was 20.4 ± 2.2 years [Me: 20, IQR: 19 - 21] and ranged from 18 to 27 years. No statistically significant difference in age was found when comparing the absence and presence of orthodontic appliances (metal and non-metal) (p=0.9819).

There was no statistically significant difference between groups in relation to sex and socioeconomic status, as shown in Table 1. Middle socioeconomic status was the most frequent in all groups (p=0.594). Regarding oral hygiene, the median for tooth brushing was three times a day for all three groups, the median for use of dental floss was once a day, which corresponded to the group with metal

Table 1. Frequency distribution of sex and socioeconomic status in the appliance-free, metal appliance, and non-metal appliance groups.

Variable	Appliance-free n (%)	Metal Ap. n (%)	Non-metal Ap. n (%)	P
Sex				0.381
Female	3 (42.9)	5 (71.4)	6 (85.7)	
Male	4 (57.1)	2 (28.6)	1 (14.3)	
Socioeconomic status				0.594
Low	1 (14.3)	3 (42.9)	1 (14.3)	
Middle	4 (57.1)	4 (57.1)	5 (71.4)	
High	2 (28.6)	0 (0.0)	1 (14.3)	

Fisher's Exact test. Ap.: Appliances.

Table 2. Median and interquartile range of daily oral hygiene measures reported in the different groups.

Oral hygiene measure	Appliance-free Me (IQR)	Metal Ap. Me (IQR)	Non-metal Ap. Me (IQR)	P
Tooth brushing F.	3 (3 - 3)	3 (2 - 3)	3 (2 - 3)	0.8465
Use of dental floss F.	0 (0 - 1)	1 (0 - 1)	0 (0 - 1)	0.3189
Use of mouthwash F.	1 (0 - 1)	1 (0 - 2)	1 (1 - 2)	0.3189

Kruskal-Wallis test. F.: Frequency per day. Me: Median. IQR: Interquartile range.

Table 3. Median and interquartile range of salivary pH and salivary *S. mutans* CFU/mL count in the appliance-free, metal appliance, and non-metal appliance groups.

Variable	Appliance-free Me (IQR)	Metal Ap. Me (IQR)	Non-metal App. Me (IQR)	P
Salivary pH	7.6 (7.3 - 7.8)	7.3 (7.0 - 7.4)	7.6 (7.6 - 7.8)	**0.0478**
CFU/mL count ($\times 10^3$)	56.0 (9.2 - 75.5)	28.6 (9.6 - 64.0)	18.6 (9.6 - 67.0)	0.7459

Kruskal-Wallis test. CFU: Colony-forming Units. Me: Median. IQR: Interquartile range.

appliances, and the median for use of mouthwash was once a day for all groups (Table 2).

In relation to salivary pH, the median was lower [Me: 7.3, IQR: 7.0 - 7.4] in the group with metal appliances than in the other groups, with a statistically significant difference (p=0.0478). Furthermore, statistically significant differences were found between not having any orthodontic appliance and having metal appliances (p=0.0239), and between having metal and non-metal appliances (p=0.0118) (Table 3).

For *S. mutans* CFU/mL count, no statistically significant difference was observed (p=0.7459), although the median count was lower in the two groups with orthodontic appliances than in the appliance-free group (Table 3, Fig. 1).

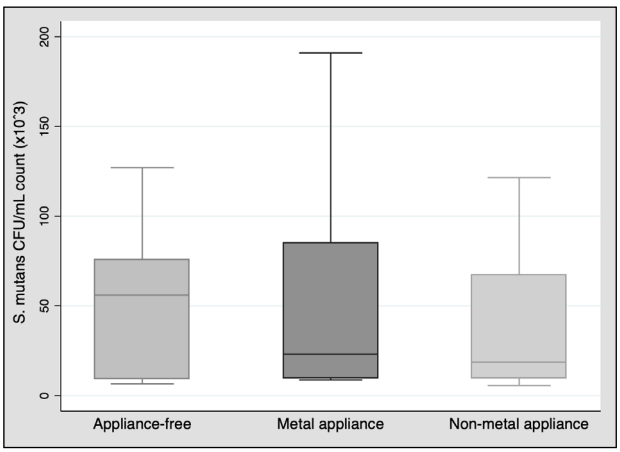

Fig. 1: *Salivary S. mutans CFU/mL count in the three groups.*

DISCUSSION

The *S. mutans* CFU/mL count did not differ significantly between groups (appliance-free, metal appliances and non-metal appliances). Nonetheless, a statistically significant difference was found in saliva pH levels between the group without appliances and the group with metal appliances, as well as between the groups with metal and non-metal appliances, possibly due to lower saliva pH levels in the group with metal appliances.

Papaioannou et al.[13] conducted an in vitro investigation to evaluate the adhesion of *S. mutans* to the surfaces of three types of brackets (six metal, six ceramic, and six plastic) covered with a salivary pellicle. Although a higher number of *S. mutans* CFU was found on metal brackets, the difference was not statistically significant (p=0.360). The authors suggest that the affinity of *S. mutans* to one material or another cannot be confirmed because it is the salivary properties that influence this adhesion. They also mention that *Streptococcus sanguis* may have an antagonistic relationship with *S. mutans*, possibly preventing its adhesion[13]. In the present study, the salivary *S. mutans* CFU/mL count was higher in the group with metal appliances [Me: 28.6×10^3] than in the group with non-metal appliances [Me: 18.6×10^3] but the difference was not statistically significant (p=0.8480).

Similarly, in another study, Jurela et al. found no statistically significant difference between salivary *S. mutans* CFU/mL counts in participants with fixed metal and non-metal appliances, even though the number of colonies was higher in participants with metal appliances[17], as was the case in the present investigation.

Another factor that affects salivary pH and biofilm organization is the host's diet[5]; though it has been challenging to understand the complexity of microbial communities interacting with the nutrients in the diet[18]. The authors of a recent systematic review concluded that fixed orthodontic appliances cause major changes in the oral microbiota one month after being installed, regardless of the difficulty of standardizing the results given the different methods for sample collection and microbial count. None of the 51 prospective studies assessed the patients' diet, and not all of them presented high methodological quality[4].

A review by Freitas et al. (2014) found moderate evidence supporting changes in the quantity and quality of the oral microbiota due to the presence of fixed orthodontic appliances, possibly because it included eight articles with samples collected from oral mucosa as well as intraoral appliances. The review also highlights the lack of diet assessment and the impact of individual oral hygiene practices[19]. In our study, all three groups reported good oral hygiene measures, including tooth brushing, use of dental floss and mouthwash, perhaps because all the participants were university students, of whom 13 (61.9%) were in the undergraduate dentistry degree program. These 13 participants were evenly distributed across the three groups.

Regarding salivary pH levels, previous studies recommend caution when reviewing study results because salivary properties may vary depending on age, circadian rhythm, time of day and level of hydration, among other factors[20]. In the current study, salivary pH was measured using a digital pH meter, and all samples were collected during the morning, allowing no more than 30 minutes between collection and measurement. Lower pH values were found in the group with metal orthodontic appliances (p=0.0478), as was also reported by Kanaya et al., who found lower salivary pH levels in participants with orthodontic appliances than in those without orthodontic appliances (p=0.001)[20].

The morning was chosen as the best time to collect the saliva because many studies reported having conducted this procedure between 7:30 and 11:30 a.m.[21,22,23]. These studies suggest that collecting saliva samples in the morning helps control fluctuations in salivary microbial counts that occur throughout the day. Petti et al. (2001) also recommend that saliva samples should be collected before eating and tooth brushing[24], as it was done in the current study.

A limitation of the current study was the sample size. There were 21 participants, in accordance with sample sizes reported in previous studies with similar aims, albeit with different designs. The cross-sectional design used is also a limitation because saliva samples were collected at one specific point in time instead of longitudinally. Another limitation was not having carried out a clinical examination, even though the study did not aim to correlate oral health status with the *S. mutans* CFU/mL count.

Despite these limitations, it is important to note that this study estimated microbial populations using saliva samples to count *S. mutans* CFU/mL, which involves more methodical and rigorous labor than do microbial

counts using bacteria test kits. Moreover, there are advantages to using saliva samples: saliva is easy to collect, the procedure is non-invasive, and there is no requirement to collect saliva from specific sites within the oral cavity, unlike bacterial plaque samples, which are generally site-specific[25]. Nonetheless, excellent agreement has been found between results from stimulated saliva samples and plaque samples for quantitative evaluation of *S. mutans*[26].

This study serves as a reference point for further studies on salivary *S. mutans* CFU count in relation to the presence and absence of metal and non-metal orthodontic appliances, considering that there are currently no publications on the subject in Colombia.

CONCLUSIONS

No statistically significant difference in salivary *S. mutans* CFU/mL count was found between groups. Salivary pH was lower in the group with metal appliances than in the groups with non-metal appliances and without appliances.

DECLARATION OF CONFLICTING INTERESTS

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Marda A, Elhamzaoui S, El Mansari A, Souly K, Farissi F, Zouhdi M, et al. Evaluation of changes in cariogenic bacteria in a young Moroccan population with fixed orthodontic appliances. Int J Dent. 2018;2018:5939015. https://doi.org/10.1155/2018/5939015
2. Maret D, Marchal-Sixou C, Vergnes JN, Hamel O, Georgelin-Gurgel M, Van Der Sluis L, et al. Effect of fixed orthodontic appliances on salivary microbial parameters at 6 months: a controlled observational study. J Appl Oral Sci. 2014;22(1):38-43. https://doi.org/10.1590/1678-775720130318
3. Metin-Gürsoy G, Uzuner FD. The Relationship between Orthodontic Treatment and Dental Caries. In: Akarslan Z, ed. *Dental Caries - Diagnosis, Prevention and Management*: IntechOpen; 2018:55-77.
4. Lucchese A, Bondemark L, Marcolina M, Manuelli M. Changes in oral microbiota due to orthodontic appliances: a systematic review. J Oral Microbiol. 2018;10(1):1476645. https://doi.org/10.1080/20002297.2018.1476645
5. Meyer-Lueckel H, Paris S, Ekstrand KR. Caries Management - Science and Clinical Practice. Stuttgard: Thieme; 2013.
6. Quirynen M, Brecx M, van Steenberghe D. Biofilms in the Oral Cavity: impact of Surface Characteristics. In: Evans L, ed. *Biofilms: Recent Advances in their Study and Control*. Amsterdam: Harwood Academic Publishers; 2005:173-194.
7. Lee SP, Lee SJ, Lim BS, Ahn SJ. Surface characteristics of orthodontic materials and their effects on adhesion of mutans streptococci. Angle Orthod. 2009;79(2):353-360. https://doi.org/10.2319/021308-88.1
8. Eliades T, Eliades G, Brantley WA. Microbial attachment on orthodontic appliances: I. Wettability and early pellicle formation on bracket materials. Am J Orthod Dentofacial Orthop. 1995;108(4):351-360. https://doi.org/10.1016/s0889-5406(95)70032-3
9. Sundararaj D, Venkatachalapathy S, Tandon A, Pereira A. Critical evaluation of incidence and prevalence of white spot lesions during fixed orthodontic appliance treatment: A meta-analysis. J Int Soc Prev Community Dent. 2015;5(6):433-439. https://doi.org/10.4103/2231-0762.167719
10. Alessandri Bonetti G, Incerti Parenti S, Garulli G, Gatto MR, Checchi L. Effect of fixed orthodontic appliances on salivary properties. Prog Orthod. 2013;14:13. https://doi.org/10.1186/2196-1042-14-13
11. Arab S, Nouhzadeh Malekshah S, Abouei Mehrizi E, Ebrahimi Khanghah A, Naseh R, Imani MM. Effect of Fixed Orthodontic Treatment on Salivary Flow, pH and Microbial Count. J Dent (Tehran). 2016;13(1):18-22.
12. Brusca MI, Chara O, Sterin-Borda L, Rosa AC. Influence of different orthodontic brackets on adherence of microorganisms in vitro. Angle Orthod. 2007;77(2):331-336. https://doi.org/10.2319/0003-3219(2007)077[0331:IODOBO]2.0.CO;2
13. Papaioannou W, Gizani S, Nassika M, Kontou E, Nakou M. Adhesion of Streptococcus mutans to different types of brackets. Angle Orthod. 2007;77(6):1090-1095. https://doi.org/10.2319/091706-375.1
14. Choi JE, Lyons KM, Kieser JA, Waddell NJ. Diurnal variation of intraoral pH and temperature. BDJ Open. 2017;3:17015. https://doi.org/10.1038/bdjopen.2017.15
15. Gold OG, Jordan HV, Van Houte J. A selective medium for Streptococcus mutans. Arch Oral Biol. 1973;18(11):1357-1364. https://doi.org/10.1016/0003-9969(73)90109-x
16. República de Colombia, Ministerio de Salud. Resolución No. 08430 de 4 de octubre de 1993. Bogotá1993.
17. Jurela A, Repic D, Pejda S, Juric H, Vidakovic R, Matic I et al. The effect of two different bracket types on the salivary levels of S mutans and S sobrinus in the early phase of orthodontic treatment. Angle Orthod. 2013;83(1):140-145. https://doi.org/10.2319/030612-187.1
18. Florez Salamanca EJ, Dantas RM, Rodriguez MJ, Klein MI. Establishment of microcosm biofilm models that reproduce a cariogenic diet intake. Biofouling. 2020;36(10):1196-1209. https://doi.org/10.1080/08927014.2020.1862093
19. Freitas AO, Marquezan M, Nojima Mda C, Alviano DS, Maia LC. The influence of orthodontic fixed appliances

on the oral microbiota: a systematic review. Dental Press J Orthod. 2014;19(2):46-55. https://doi.org/10.1590/2176-9451.19.2.046-055.oar

20. Kanaya T, Kaneko N, Amaike C, Fukushima M, Morita S, Miyazaki H, et al. A study on changes in caries risk and microbial flora with the placement of edgewise appliance. Orthodontic Waves. 2007;66:27-32. https://doi.org/10.1016/j.odw.2007.03.001
21. Aminabadi NA, Erfanparast L, Ebrahimi A, Oskouei SG. Effect of chlorhexidine pretreatment on the stability of salivary lactobacilli probiotic in six- to twelve-year-old children: a randomized controlled trial. Caries Res. 2011;45(2):148-154. https://doi.org/10.1159/000325741
22. Caglar E, Kavaloglu SC, Kuscu OO, Sandalli N, Holgerson PL, Twetman S. Effect of chewing gums containing xylitol or probiotic bacteria on salivary mutans streptococci and lactobacilli. Clin Oral Investig. 2007;11(4):425-429. https://doi.org/10.1007/s00784-007-0129-9
23. Cortes-Dorantes N, Ruiz-Rodriguez MS, Karakowsky-Kleiman L, Garrocho-Rangel JA, Sanchez-Vargas LO, Pozos-Guillen AJ. Probiotics and their effect on oral bacteria count in children: a pilot study. Eur J Paediatr Dent. 2015;16(1):56-60.
24. Petti S, Tarsitani G, D'Arca AS. A randomized clinical trial of the effect of yoghurt on the human salivary microflora. Arch Oral Biol. 2001;46(8):705-712. https://doi.org/10.1016/s0003-9969(01)00033-4
25. Jung WS, Kim H, Park SY, Cho EJ, Ahn SJ. Quantitative analysis of changes in salivary mutans streptococci after orthodontic treatment. Am J Orthod Dentofacial Orthop. 2014;145(5):603-609. https://doi.org/10.1016/j.ajodo.2013.12.025
26. Dasanayake AP, Caufield PW, Cutter GR, Roseman JM, Kohler B. Differences in the detection and enumeration of mutans streptococci due to differences in methods. Arch Oral Biol. 1995;40(4):345-351. https://doi.org/10.1016/0003-9969(94)00164-7

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Periodontitis prevalence and associated factors: a comparison of two examination protocols

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ABSTRACT

The fact that there are different epidemiological definitions of periodontitis and different evaluation protocols affects the estimate of periodontitis prevalence and of the influence of associated factors. The gold standard for periodontal examination is full-mouth record assessing CAL and PD. However, there are not always sufficient human and financial resources available to apply such assessment for epidemiological surveillance systems. ***Aim:*** *This study was conducted to compare different protocols and definitions of periodontitis for assessing prevalence and the impact of related factors in adult patients who requested care at the School of Dentistry, UdelaR.* ***Materials and Method:*** *This was a cross-sectional study of 410 subjects with a high burden of disease in terms of NCDs and periodontitis. Clinical examination evaluated PD in all teeth and CAL in the CPI sextants (WHO 2013). Four periodontitis criteria were defined based on two examination protocols (WHO 2013 and WHO 1997) and two definitions of epidemiological case. Comparisons were made taking the 2013 WHO protocol as a reference.* ***Results:*** *Comparison of the two examination protocols showed that prevalence was underestimated when the WHO 1997 protocol was used to define moderate-severe and severe periodontitis, by 20% and 60%, respectively.* ***Conclusions:*** *When the severity of periodontitis was not considered, the WHO 2013 protocol did not provide more information on what factors increase the chance of periodontitis. However, when severity was considered, the associated factors were different. Consequently, in a small population, it would be worth using the WHO 2013 protocol, which is the closest to the full-mouth gold standard criterion.*

Keywords: *cross-sectional studies - periodontal index - periodontitis - risk factors - non-communicable diseases*

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Prevalencia y factores asociados a periodontitis: comparación de dos protocolos de examen WHO

RESUMEN

La variabilidad en la definición epidemiológica de la periodontitis y los protocolos de evaluación afectan la medición de la prevalencia y su asociación con ciertos factores. Si bien, el patrón oro para el examen periodontal es el registro de boca completa, que evalúa la pérdida de inserción (CAL, por sus siglas en inglés) y profundidad de sondaje (PS, por sus siglas en inglés), los recursos no siempre están disponibles para los sistemas de vigilancia epidemiológica. ***Objetivo:*** *En este estudio se compararon diferentes protocolos y definiciones de periodontitis evaluando la prevalencia y la asociación de factores relacionados en pacientes adultos que solicitaron atención en la Facultad de Odontología de la UdelaR.* ***Materiales y Método:*** *Los datos provienen de un estudio transversal de 410 sujetos con una elevada carga de enfermedad en términos de ENT y periodontitis. Se utilizó un examen clínico registrando PD en todos los dientes y CAL en los sextantes CPI (WHO 2013). Se definieron cuatro criterios de periodontitis basados en dos protocolos de examen y dos definiciones epidemiológicas de caso. Las comparaciones se realizaron tomando como referencia el protocolo de la OMS de 2013.* ***Resultados:*** *Al comparar los dos protocolos de examen, se verificó la subestimación de la prevalencia cuando se utilizó el protocolo de la OMS de 1997 para la definición de caso moderado-grave y para la periodontitis grave, siendo en el primer caso del 20% y en el segundo caso tres veces mayor que.* ***Conclusiones:*** *Si no se considera la gravedad de la periodontitis, el uso del protocolo de la OMS 2013 no proporciona más información sobre qué factores aumentan la probabilidad de periodontitis. Sin embargo, al analizar la gravedad, los factores asociados fueron diferentes. En consecuencia, en una población pequeña estaría justificado el esfuerzo de utilizar el protocolo de la OMS de 2013, ya que el criterio de referencia es la boca completa.*

Palabras clave: *estudios transversales - índice periodontal - periodontitis - factores de riesgo - enfermedades no transmisibles*

INTRODUCTION

Periodontal disease is a public health problem that affects numerous global populations. It is the leading cause of tooth loss, particularly in older adults, significantly affecting their quality of life[1]. It is a multifactorial, inflammatory-based, socio-culturally modeled disease[2], like other chronic noncommunicable diseases (NCDs). Its prevalence ranges from 15 to 47%[1,2] worldwide, and 10.8% of the population is affected by severe periodontitis. Brazil's 2010 population survey showed that the prevalence of "moderate to severe" periodontal disease in adults aged 35-44 years was 15.3%[3]. In Uruguay, Lorenzo et al. reported that 22% of the population over 35 years of age presented moderate-severe forms of periodontitis, and that 9% presented the severe form[4]. Several factors associated to periodontitis have been described: age, sex and socioeconomic status; tobacco consumption[5], and with a lower level of evidence, the consumption of acohol[6], the intake of fruits and vegetables[7,8], and as associated pathologies, diabetes[9] and obesity[10]. Different definitions and examination protocols have been used to study prevalence, severity and the associations mentioned, to the point that the epidemiological definition of "periodontitis case" has become extremely controversial[11–14].

The use of different definitions has decisive influence on the epidemiological surveillance of periodontitis by comparing results of prevalence and severity[12-15], as well as on the direction and magnitude of the associations with the risk factors and systemic diseases analyzed[16]. The gold standard of periodontal examination is the full-mouth periodontal examination (FMPE) protocol based on the clinical examination of 6 sites per tooth, including the study of clinical attachment loss (CAL) and probing depth (PD) as continuous variables[17]. However, the time, logistics and costs involved in applying FMPE sometimes render it impractical for epidemiological surveillance systems[18,19]. To compensate for these difficulties, different partial mouth periodontal examination (PMPE) protocols have been developed, which examine certain teeth [Community Periodontal index (CPI), Ramfjord], randomly selected quadrants, among others[20]. These protocols have demonstrated varying degrees of accuracy compared to FMPE. They underestimate the prevalence, and sometimes overestimate the severity of the pathology, the dental sector, and the population age[3,21,22].

One of the partial protocols widely used in public health, especially in populational oral health surveys, is the Community Periodontal Index (CPI)[23] proposed by the WHO in the 1990s. The CPI divides the mouth into sextants and uses index teeth to evaluate periodontitis. This indicator has been modified and improved over time. For example, national oral health studies in Brazil have adopted a recording system in which the prevalence of each condition can be counted per sextant, not recording only the worst condition[24]. In 2013, the WHO developed a new manual for basic national surveys, in which it introduced the full mouth examination to assess probing depth (PD), while maintaining the use of six sextants for the clinical attachment loss (CAL) examination[25]. The CPI, with several of the aforementioned strengths and weaknesses, is still used for population surveys, creating the need to evaluate its new version.

In order to evaluate the weaknesses and strengths of each method, the aim of this article was to compare different protocols and definitions of periodontitis by assessing the prevalence and the magnitude of association of related factors in adult patients who requested care at the Screening and Admission Service of the School of Dentistry, UdelaR (Uruguay).

MATERIALS AND METHOD

Study design and sample

This was a cross-sectional, observational study conducted at the School of Dentistry (F.O)/ UdelaR. Participants were patients who requested care at the institution's Screening and Admission Service. The study was conducted from August 2015 to December 2016.

A representative sample was used. Over a 6-month period, the Screening and Admissions Service receives 4000 people requesting dental care. The sample was planned to obtain prevalence of up to 30% in two estimation domains with a 95% confidence level and a 5% margin of error. Sample size was estimated as 620 patients. The eligibility criteria were being older than 18 years, and having requested dental care at the Screening and Admission Service from August to December 2015 or March to May 2016. For the present study, considering epidemiological characteristics of periodontitis affecting adults and elderly adults, patients younger than 35 years (n=210) were excluded.

Data collection

Dental examination was performed in a dental chair, with mirror, clamp and CPI probe, recording PD in all teeth present[25] and CAL per sextant. Each tooth was probed at six sites (mesio-buccal, mid-buccal, disto-buccal, disto-lingual, mid-lingual and mesio-lingual). PD was measured as the distance from the free gingival margin to the base of the gingival crevice, and subjects were classified using the following categorization: "absent", when pocket depth was 0 to 3 mm, and "present" when pocket depth was ≥ 4mm. Edentulous subjects were not considered. The Brazilian national oral health survey modification for CPI was used. It consisted of measuring periodontal conditions (PD and CAL) on each tooth and reporting the value for each condition, not only the worst situation as the original CPI index does.

Participating examiners were calibrated by means of the examination of 30 volunteers covering all the age ranges to be included in the study. The kappa coefficient was calculated for the CPI indicator, and PD and CAL were calibrated, obtaining values of 0.71 and 0.79, respectively.

The participants answered a questionnaire on socio-demographics (sex, age, studies completed, monthly income and medical and dental healthcare coverage), behavior (quality of diet, alcohol and tobacco consumption, physical activity) and metabolic risk factors (history of hypertension, diabetes, high cholesterol and altered glycemia).

The question about education recorded participants' maximum educational level; the question about income recorded average monthly household income (in Uruguayan pesos), and the question about health and dental coverage recorded participants' healthcare providers.

The questions about diet included the amount and frequency of weekly consumption of fruits and vegetables; sugary drinks and products; ready-to-eat, high-salt products; use of salt when cooking and eating, and type and frequency of alcohol consumption. The questions about smoking enquired about whether the participant smoked and if so, how much. The questions about physical activity considered frequency and duration of moderate and intense physical activity, using a chart with examples[26].

Glycemia was measured by capillary puncture using disposable lancets and test strips for measurement on a digital glucometer (Bayer® Contour TS), following the manufacturer's recommendations. Blood pressure (BP) was measured with an aneroid sphygmomanometer (RIESTER® verified by LATU No. 333075. 06/2015), after the patient had sat at rest for 15 minutes, with his/her arm relaxed on a firm surface, using an appropriately sized cuff for arm perimeter, and recording a single measurement

Anthropometric data were height, weight and waist circumference. Body height (Seca®) was measured to the nearest 0.5 cm. Weight (Seca®) was recorded to the nearest 100 grams with patients wearing light indoor clothing without shoes. Waist circumference was measured with an inelastic measuring tape (Seca®), following the WHO protocol[25].

Variables and indicators

Dental variables

Four periodontitis criteria were defined based on two examination protocols and two epidemiological definitions of periodontitis, combined as follows (Fig. 1).

- *Partial protocol, moderate-severe*: examination of CPI sextants with their index teeth, considering

	PROTOCOLS					
	WHO 1997		WHO 2013		CASE DEFINITION OF PD	
	PP	CAL	PP	CAL	moderate-severe	severe
Partial protocol, moderate-severe	CPI sextants	CPI sextants			X	
Partial protocol, severe	CPI sextants	CPI sextants				X
Full mouth, moderate-severe			All teeth	CPI sextants	X	
Full mouth, severe			All teeth	CPI sextants		X

Fig. 1: *Case Definition of Periodontitis*

"moderate and severe" disease: moderate to severe when CPI > 2 (periodontal pocket ≥ 4mm and CAL ≥ 4mm).

- *Partial protocol, severe*: examination of sextants with their index teeth, considering severe periodontitis when CPI > 2 (periodontal pocket ≥ 4mm and CAL ≥ 6mm).
- *Full mouth, moderate-severe:* complete mouth examination for PD and by sextant for CAL, considering "moderate to severe" periodontitis as in item 1.
- *Full mouth, severe:* complete mouth examination for PD and by sextant for CAL, considering "severe" periodontitis as in item 2.

Socio-demographic variables

Educational level was classified as primary education or lower; secondary education or a lower; and tertiary education or higher. Health coverage was classified as either public or private coverage. Income level was classified as up to 700 USD, from 700 to 1000 USD, from 1000 to 1200 USD, or higher than 1200 (according to the local dollar exchange rate in 2015).

Behavioral variables

Dietary risk factors were considered to be present when the consumption of fruits and vegetables was < 5 servings per day; the consumption of sugary drinks, sugary products and ready-to-eat foods was > 2 times a week; the addition of salt to ready-to-eat preparations (considering the answers "always/almost always"). The alcohol consumption variable considered the frequency and number of drinks, with the categories: "does not consume", "consumes" and "harmful consumption". Consumption was considered harmful when respondents reported having at least 5 drinks (on one of those days) or drinking alcohol more than twice a week. For tobacco, 3 categories were considered: never, currently or formerly consumed. Insufficient physical activity was considered to be less than 75 minutes per week of vigorous physical activity or less than 150 minutes per week of moderate physical activity.

Metabolic variables

Hypertension was considered as systolic BP greater than or equal to 140 mmHg and/or diastolic BP greater than or equal to 90 mmHg[27], or self-reported hypertension. Diabetes was defined as capillary glycemia ≥ 200 mg/dl with or without fasting, or a self-reported history of diabetes diagnosed by respondent's physician[26]. Obesity and overweight were analyzed through body mass index [BMI = weight (kg)/square of height (m^2)] and considered as BMI values of 25.0 to 29.9 kg/m^2 and 30 kg/m^2 or more, respectively. Altered values of waist circumference corresponded to > 88 cm in women and > 102 cm in men.

Ethical aspects

This study was approved by an Institutional Review Board (Uruguayan Research Ethics Committee) of the School of Dentistry (F.O; file number 091900-000121-14) and was conducted in accordance with the Helsinki Declaration of 1975, as revised in 2013. Participation was voluntary, and participants signed a written informed consent. The participants were informed of the results of the measurements (weight, height, waist circumference, capillary glycemia and blood pressure) and advised on the need for medical consultation when appropriate.

Statistical analysis

After performing the descriptive analysis, 4 logistic regression models were adjusted, one for each periodontitis criterion. These models included explanatory variables in hierarchical blocks. First, sociodemographic factors were included, then metabolic variables and lastly, factors referring to behavioral variables. The association was described using odds ratios (OR), assessing its uncertainty using 95% confidence intervals. All statistical procedures were performed in the R[28] software.

RESULTS

A total 410 subjects were analyzed. Table 1 provides the data recorded. The population was predominantly female (58.8%); with 79.0% in the 35- to 64-year age range, and 16% having university studies. Regarding income, 58.2% earned less than 700 USD, and only 9% more than 1200 USD. Considering oral health, 70% used the private subsector, and 9.8% used the public subsector of the National Integrated Health System. Table 1 reports prevalence of NCDs and their risk factors.

The prevalence of periodontitis according to the 4 criteria used to assess it in the study population was: "Partial protocol, moderate-severe": 43.9%

Table 1. Characteristics of the study population

Sociodemograhic	n	%	
Sex			
Female	241	58,8%	(54% - 63,5%)
Male	169	41,2%	(36,5% - 46%)
Age			
35 - 44	114	27,8%	(23,5% - 32,1%)
45 - 64	210	51,2%	(46,4% - 56,1%)
65 - 74	59	14,4%	(11% - 17,8%)
≥ 74	27	6,6%	(4,2% - 9%)
Healthcare system			
ASSE	264	64,4%	(59,8% - 69%)
Mutualist	131	32,0%	(27,4% - 36,5%)
Other	15	3,7%	(1,8% - 5,5%)
Dental care			
ASSE	287	70,0%	(65,6% - 74,4%)
Mutualist	40	9,8%	(6,9% - 12,6%)
Others	83	20,2%	(16,4% - 24,1%)
Income (thousands)			
≤ 17	228	58,2%	(53,4% - 62,9%)
17 - 24	76	19,4%	(15,6% - 23,2%)
24 - 32	51	13,0%	(9,8% - 16,3%)
≥ 32	37	9,4%	(6,6% - 12,3%)
Education			
primary (≤6 years)	134	32,7%	(28,1% - 37,2%)
secondary (6 - 12 years)	210	51,2%	(46,4% - 56,1%)
tertiary (≥12 years)	66	16,1%	(12,5% - 19,7%)
NCD			
Hypertension			
sick	216	52,7%	(47,9% - 57,5%)
heathy	194	47,3%	(42,5% - 52,1%)
Obesity			
sick	270	65,9%	(61,3% - 70,4%)
healthy	140	34,1%	(29,6% - 38,7%)
Diabetes			
sick	77	18,8%	(15% - 22,6%)
healthy	333	81,2%	(77,4% - 85%)
Habits and consumption			
Harmful alcohol consumption			
does not drink	171	41,7%	(36,9% - 46,5%)
no	209	51,0%	(46,1% - 55,8%)
yes	30	7,3%	(4,8% - 9,8%)
Physical activity			
insufficient	196	47,8%	(43% - 52,6%)
sufficient	214	52,2%	(47,4% - 57%)
Always or almost always adds salt			
yes	60	14,6%	(11,2% - 18,1%)
no	350	85,4%	(81,9% - 88,8%)
Servings of fruits and vegetables			
5 or more	39	9,5%	(6,7% - 12,4%)
fewer than 5	371	90,5%	(87,6% - 93,3%)
Consumption of sugary drinks			
2 or more days a week	193	48,9%	(44% - 53,7%)
less than 2 days a week	202	51,1%	(46,3% - 56%)
Consumption of tobacco			
currently	122	29,8%	(25,4% - 34,3%)
former	107	26,2%	(21,9% - 30,4%)
never	180	44,0%	(39,2% - 48,8%)

(95% CI: 34.8-53.9), "Partial protocol, severe": 6.5% (95% CI: 3.2-12.9), "Full mouth, moderate-severe" 53.9% (95% CI: 44.5-63.1) and "Full mouth, severe": 16.1% (95% CI: 10.3-24.2). Thus, considering as a reference the WHO 2013 protocol[25] in its two versions of periodontitis ("Full mouth, moderate-severe", and "Full mouth, severe") and comparing it to the WHO protocol 1997[23] ("Partial protocol, moderate-severe", "Partial protocol, severe"), the differential relative gap resulted in an underestimation of the disease by approximately 20% and 60% for moderate to severe and severe disease, respectively.

The result of the multivariate analysis using four models (*Model 1: Moderate and severe PD (teeth index); Model 2: Severe PD (teeth index); Model 3: Moderate and severe PD (teeth index + WHO); Model 4: Severe PD (teeth index + WHO);* each model corresponds to a criterion, is presented in Table 2. According to model 1, those most likely to have periodontitis were men [OR 1.47 (95% CI: 1.16-1.86)], people aged 45-64 years [OR 1.48 (95% CI: 1.1-2.02)], people aged 65-74 years [OR 1.99 (95% CI: 1.36-2.92)], and ex-smokers [OR 1.43 (95% CI: 1.06-1.93)]. A linear trend was observed in age, where the older the subject, the greater the chance of periodontitis (p=0.039).

In model 2, age 45-64 and 65-74 years, and a monthly income level of less than 700 USD increased the chance of presenting periodontitis, with OR 4.03 (95% CI: 1.38–17.1), 6.69 (95% CI: 1.96–30.6) and 4.21 (95% CI:1.22–26.5), respectively. Considering behavioral variables, alcohol intake and the addition

Table 2. Adjusted OR (95%) of Periodontitis in Multivariate Logistic Regression Model for different protocols and case definitions

	Moderate and severe PD (teeth index)		Severe PD (teeth index)		Moderate and severe PD (teeth index + WHO)		Severe PD (teeth index + WHO)	
	OR	IC95%	OR	IC95%	OR	IC95%	OR	IC95%
Sociodemographics								
Sex (Male)	1,474	(1,167 - 1,860)	1,661	(0,899 - 3,083)	1,878	(1,539 - 2,293)	2,548	(1,710 - 3,838)
Age (45 - 64)	1,483	(1,102 - 2,016)	4,032	(1,381 - 17,15)	1,837	(1,422 - 2,39)	3,697	(1,947 - 7,783)
Age (65 - 74)	1,999	(1,364 - 2,926)	6,695	(1,965 - 30,64)	2,504	(1,801 - 3,485)	4,858	(2,291 - 11,02)
Age (≥ 74)	1,568	(0,931 - 2,577)	3,980	(0,820 - 21,37)	1,983	(1,277 - 3,044)	3,426	(1,296 - 8,933)
Healthcare service (Mutualist)	1,098	(0,615 - 2,059)	1,055	(0,308 - 4,907)	0,787	(0,476 - 1,329)	0,683	(0,297 - 1,732)
Healthcare service (ASSE)	0,776	(0,449 - 1,418)	0,547	(0,179 - 2,387)	0,733	(0,457 - 1,202)	0,596	(0,283 - 1,417)
Dental care (Mutualist)	1,250	(0,806 - 1,968)	1,474	(0,411 - 6,897)	1,152	(0,785 - 1,704)	1,230	(0,543 - 3,055)
Dental care (ASSE)	1,202	(0,782 - 1,884)	2,225	(0,683 - 10,09)	1,005	(0,69 - 1,48)	1,412	(0,651 - 3,432)
Education (primary)	0,799	(0,558 - 1,151)	0,386	(0,160 - 0,918)	0,803	(0,592 - 1,091)	0,523	(0,302 - 0,907)
Education (secondary)	1,029	(0,747 - 1,433)	0,572	(0,276 - 1,236)	0,955	(0,725 - 1,265)	0,620	(0,381 - 1,027)
Income (less than 17)	1,147	(0,772 - 1,745)	4,207	(1,219 - 26,532)	1,238	(0,876 - 1,774)	1,955	(0,985 - 4,349)
Income (17 - 24)	1,153	(0,739 - 1,827)	3,293	(0,832 - 21,926)	1,055	(0,714 - 1,571)	1,468	(0,669 - 3,483)
Income (24 - 32)	1,090	(0,671 - 1,785)	0,458	(0,021 - 4,905)	1,229	(0,807 - 1,882)	0,893	(0,322 - 2,440)
NCD								
HTN (sick)	1,091	(0,845 - 1,409)	1,191	(0,608 - 2,395)	0,990	(0,797 - 1,23)	0,962	(0,627 - 1,483)
BMI (overweight/obesity)	1,012	(0,777 - 1,324)	0,588	(0,302 - 1,166)	0,925	(0,74 - 1,159)	0,817	(0,532 - 1,270)
Diabetes (sick)	1,073	(0,793 - 1,438)	0,982	(0,427 - 2,049)	1,127	(0,871 - 1,451)	0,914	(0,536 - 1,496)
Habits and consumption								
Harmful consumption of alcohol (no)	1,160	(0,894 - 1,509)	0,923	(0,450 - 1,916)	1,155	(0,923 - 1,447)	0,919	(0,584 - 1,451)
Harmful consumption of alcohol (yes)	1,001	(0,604 - 1,615)	3,258	(1,133 - 8,914)	1,216	(0,808 - 1,812)	1,571	(0,774 - 3,080)
Physical activity (insufficient)	1,025	(0,798 - 1,316)	1,547	(0,799 - 3,052)	1,043	(0,841 - 1,293)	1,297	(0,847 - 1,995)
Added salt SCS (yes)	1,029	(0,721 - 1,444)	2,179	(1,030 - 4,604)	1,008	(0,746 - 1,349)	1,467	(0,842 - 2,461)
Consumption of fruits and vegetables (less than 5)	1,207	(0,804 - 1,781)	2,294	(0,892 - 5,426)	1,301	(0,918 - 1,826)	2,027	(1,137 - 3,504)
Sugar consumption (more than 2 days a week)	0,985	(0,76 - 1,276)	0,815	(0,397 - 1,623)	1,003	(0,803 - 1,252)	0,661	(0,421 - 1,024)
Tobacco consumption (current)	1,253	(0,916 - 1,709)	0,447	(0,173 - 1,058)	1,593	(1,224 - 2,074)	0,920	(0,536 - 1,556)
Tobacco consumption (former)	1,433	(1,062 - 1,933)	0,929	(0,427 - 1,957)	1,349	(1,038 - 1,752)	0,877	(0,526 - 1,448)

of salt in ready-to-eat foods constituted risk factors with OR 3.26 (95% CI: 1.13–8.91) and 2.18 (95% CI: 1.03 – 4.60), respectively. Having only primary studies decreased the chance of presenting periodontitis [OR 0.39 (95% CI: 0.16 – 0.92)]. The presence of a linear trend in the educational level was observed, where the higher the subject's level of study, the more likely they were to report periodontitis (p=0.031). It was also observed that the lower the income, the more chances of periodontitis (p=0.012).

In model 3, the factors that increased the chance of periodontitis were: male sex [OR 1.88 (95% CI:1.54-2.29)], age 45-64 years or 65 -74 years [OR 1.84 (95% CI:1.42 -2.4)] and OR 2.50 (95% CI:1.80-3.49)], respectively, and current consumption [OR 1.59 (95% CI:1.22-2.07)] or past consumption [OR 1.35 (95% CI:1.04-1.75)] of tobacco. The presence

of a linear trend in age was observed, where the older the subject, the greater the chance of periodontitis (p<0.001).

Finally, in model 4, the factors that increased the chance of periodontitis were: male sex [OR 2.55 (95% CI: 1.71–3.84)], age 45-64 years or 65-74 years [OR 3.70 (95% CI: 1.95-7.78)] and [OR 4.86 (95% CI:2.29-11.02)], respectively, and consuming < 5 servings of fruits and vegetables daily [OR 2.03 (95% CI: 1.14-3.50)]. Having only primary education was a protective factor [OR 0.53 (95% CI: 0.30-0.90)]. The presence of a linear trend in age was observed, where the older the age, the greater the chances of periodontitis (p=0.007). It was also observed that the higher the educational level, the more chances of periodontitis (p=0.021), and the lower the income level, the more chances of periodontitis (p=0.039).

Analysis of the models as a whole showed that age was associated in all models, and male sex in three of them. In models that refer to moderate to severe periodontitis ("Partial protocol, moderate-severe" and "Full mouth, moderate-severe"), smoking was also as a risk factor. It was observed that the effect of age as a risk factor was more pronounced in the full mouth model than in the partial mouth model (Table 2). In the "Partial protocol, severe" and "Full mouth, severe" models corresponding to severe disease, having university studies increased the chance of periodontitis.

DISCUSSION

The primary objective of the study was to compare two protocols for recording periodontitis[23,25] in relation to two case definitions of periodontitis (moderate-severe and severe). The four criteria used were "Full mouth, moderate-severe", "Full mouth, severe", "Partial protocol, moderate-severe" and "Partial protocol, severe". The comparisons were made in a population with a high burden of disease in terms of NCDs and periodontitis, and taking as a reference the WHO 2013 protocol. Comparison of the two examination protocols considered showed that prevalence was underestimated when the WHO 1997 protocol was used to define moderate-severe periodontitis and severe periodontitis, by 20% and 60%, respectively. The impact of the definition of a case of periodontitis and the way it is recorded have long been debated in the literature, generating diverse estimates of the prevalence of periodontitis[12–14]. This is one of the limitations of CPI[20,21], which is characterized by underestimation[18,29]. However, when Vettore et al.[3] studied two adult populations, they found that underestimation occurs when there are sites with incipient pockets, while in the case of moderate to severe pockets, the frequency of diseased sites was overestimated for both pockets and attachment loss. The current study found no other paper comparing different case definitions and different screening protocols at the same time.

All the criteria used showed that age was an associated factor. Sex was associated in all models except full mouth, moderate-severe. Other factors that showed association in some of the models were those related to behavior: current consumption of tobacco, harmful consumption of alcohol, addition of salt to ready-to-eat food, consumption of fruits and vegetables below the daily recommendation, and the socio-demographic variables education and income.

In the models that had moderate to severe periodontitis as a case definition, the present study showed that smoking was associated with periodontitis in both the WHO 2013 and 1997 protocols ("Partial protocol, moderate-severe" and "Full mouth, moderate-severe"). There is considerable evidence that tobacco use increases the likelihood of developing periodontitis[2,5,30–32]. It is important to note that in the study population, the proportion of smokers was 26.7%[33], which was similar to the figure reported at national level[26].

Considering severe periodontitis ("Full mouth, severe" and "Partial protocol, severe"), subjects with primary education only were less likely to have periodontitis. This differs substantially from previous studies in which the association occurs in the opposite direction (the higher the educational level, the lower the chance of periodontitis), both internationally[34,35] and nationally. This might be explained by the fact that in both models of severe disease, the population with only primary studies was older and had lost more teeth due to periodontitis. However, this would not be captured by the criterion used to measure periodontitis (PD and CAL) because it did not take into account the number of teeth lost and therefore of sextants excluded. This finding would confirm the difficulty of the CPI indicator in accounting for periodontitis in populations with a large proportion of older adults who are more likely to have lost teeth.

Another finding regarding the case definition of severe disease was that harmful consumption of alcohol tripled the chance of periodontitis in the "Full mouth, severe" model; a finding that is consistent with the international literature[36,37].

In the protocols that refer to severe disease, there are associations with less studied and diet-related risk factors, such as the consumption of salt ("Partial protocol, severe") and consumption of fruits and vegetables ("Full mouth, severe"). High salt intake has been linked to high blood pressure, an indicator of periodontitis risk. In the current study, high BP was not associated with increased chances of periodontitis, however, the addition of salt at the table was. A recent study on mice described how, compared to a high-salt diet, mice experienced increased low-impact inflammation related to hyperglycocorticoidism induced by high sodium intake. In turn, healthy volunteers who followed a salt-rich diet for a week experienced similar results[38]. These results could explain the associations found. On the other hand, adequate consumption of vegetables and fruits rich in bioactive compounds with antioxidant function has been associated with better periodontal health[39] and lower risk of periodontitis[8]. Fiber, another component of vegetables and fruits, has been associated with improvements in periodontitis markers when consumed in large quantities[40].

The current study sought to explore the association of periodontitis with various NCDs and their risk factors. The results show that there was no statistically significant association with obesity, diabetes or hypertension in the study population. These findings may be explained by the fact that this was a cross-sectional study. In addition to the diagnostic criterion, not having measured CAL in all teeth, and having applied the CPI criterion, which is less sensitive because it uses a probe that does not allow measurement in millimeters, could explain this result. Moreover, age is a factor that may interfere in the associations between the aforementioned NCDs, which are more prevalent in elderly people. Old age is also related to progressive tooth loss, leading to the exclusion of a greater number of sextants when the CPI criterion is applied.

One of the limitations of this study is the difficulty of not having the CAL record for all teeth, which would have allowed the objective of this study to be addressed more fully. The prevalence of periodontitis in Uruguay is 22%[4] (considering moderate to severe disease, WHO protocol 1997), whereas the value found in the study population is double. Working with a population with a high burden of pathology may have resulted in biases difficult to control that could affect the possibility of finding significant associations with the different factors.

Given that the CPI is recommended for epidemiological surveillance and easy to use[41], it is of interest to compare the cost-effectiveness of the 2013 and 1997 WHO protocols. According to the fieldwork conducted in the national survey of Uruguay and compared to the present work, an examination using the CPI 1997 indicator performed by a dentist who does not specialize in periodontics would take about 10 minutes, while an examination using the WHO 2013 would take about 15 minutes, i.e., one third longer. Further research should compare protocols with full-mouth CPI criteria to partial mouth, recording PD and CAL in all teeth for comprehensive evaluation of the new protocol's cost effectiveness and ability to show the effect of associated factors.

The WHO 1997 partial mouth protocol underestimated the prevalence of periodontitis when compared to that of WHO 2013 by 20% and 60% for the case definition of "moderate to severe" and "severe" periodontitis, respectively. This should be taken into account when studying populations with a high burden of disease.

To study periodontitis as a whole and its relationship with risk factors (considering both definitions of "moderate-severe" case), the complete mouth examination for periodontal pocket but not for CAL (WHO, 2013) does not provide more information on which factors increase the chance of periodontitis or on its magnitude. However, if the focus is on the definition that only includes severe cases, the associated factors expressed in the full mouth protocol (WHO 2013) and in the partial protocol (WHO 1997) are different. This would justify the effort to use the 2013 protocol, which is the closest to the full-mouth golden rule.

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DATA AVAILABILITY
The data that support the findings of this study are available upon request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

DECLARATION OF CONFLICTING INTERESTS
The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Thornton-Evans G, Eke P, Wei L, Palmer A, Moeti R, Hutchins S, Borrell LN; Centers for Disease Control and Prevention (CDC). Periodontitis among adults aged ≥30 years - United States, 2009-2010. MMWR Suppl. 2013 Nov 22;62(3):129-35.
2. Genco RJ, Borgnakke WS. Risk factors for periodontal disease. Periodontol 2000. 2013 Jun;62(1):59-94. https://doi.org/10.1111/j.1600-0757.2012.00457.x
3. Vettore MV, Lamarca Gde A, Leão AT, Sheiham A, Leal Mdo C.Partial recording protocols for periodontal disease assessment in epidemiological surveys. Cad Saude Publica. 2007 Jan;23(1):33-42. https://doi.org/10.1590/S0102-311X2007000100005
4. Lorenzo SM, Alvarez R, Andrade E, Piccardo V, Francia A, Massa F, Correa MB, Peres MA. Periodontal conditions and associated factors among adults and the elderly: findings from the first National Oral Health Survey in Uruguay. Cad Saude Publica. 2015 Nov;31(11):2425-36. https://doi.org/10.1590/0102-311X00012115
5. Thomson WM, Sheiham A, Spencer AJ. Sociobehavioral aspects of periodontal disease. Periodontol 2000. 2012 Oct;60(1):54-63. https://doi.org/10.1111/j.1600-0757.2011.00405.x
6. Amaral Cda S, Vettore MV, Leão A. The relationship of alcohol dependence and alcohol consumption with periodontitis: a systematic review. J Dent. 2009 Sep;37(9):643-51. https://doi.org/10.1016/j.jdent.2009.04.011
7. Né YGS, Martins BV, Castro MML, Alvarenga MOP et al. Is nutritional intervention an improvement factor in the management of periodontitis? A systematic review. Clin Nutr. 2020 Sep;39(9):2639-2646. https://doi.org/10.1016/j.clnu.2019.12.016
8. O'Connor JP, Milledge KL, O'Leary F, Cumming R, Eberhard J, Hirani V. Poor dietary intake of nutrients and food groups are associated with increased risk of periodontal disease among community-dwelling older adults: a systematic literature review. Nutr Rev. 2020 Feb 1;78(2):175-188. https://doi.org/10.1093/nutrit/nuz035
9. Suvan JE, Finer N, D'Aiuto F. Periodontal complications with obesity. Periodontol 2000. 2018 Oct;78(1):98-128. https://doi.org/10.1111/prd.12239
10. Chávarry NGM, Vettore MV, Sansone C, Sheiham A. The relationship between diabetes mellitus and destructive periodontal disease: a meta-analysis. Oral Health Prev Dent. 2009;7(2):107–127.
11. Beltrán-Aguilar ED, Eke PI, Thornton-Evans G, Petersen PE. Recording and surveillance systems for periodontal diseases. Periodontol 2000. 2012 Oct;60(1):40-53. https://doi.org/10.1111/j.1600-0757.2012.00446.x
12. Eke PI, Page RC, Wei L, Thornton-Evans G, Genco RJ. Update of the case definitions for population-based surveillance of periodontitis. J Periodontol. 2012 Dec;83(12):1449-54. https://doi.org/10.1902/jop.2012.110664
13. Eke PI, Dye BA, Wei L, Slade GD et al. Update on Prevalence of Periodontitis in Adults in the United States: NHANES 2009 to 2012. J Periodontol. 2015 May;86(5):611-22. https://doi.org/10.1902/jop.2015.140520
14. Tonetti MS, Claffey N; European Workshop in Periodontology group C. Advances in the progression of periodontitis and proposal of definitions of a periodontitis case and disease progression for use in risk factor research. Group C consensus report of the 5th European Workshop in Periodontology. J Clin Periodontol. 2005;32 Suppl 6:210-3. https://doi.org/10.1111/j.1600-051X.2005.00822.x
15. Savage A, Eaton KA, Moles DR, Needleman I. A systematic review of definitions of periodontitis and methods that have been used to identify this disease. J Clin Periodontol. 2009 Jun;36(6):458-67. https://doi.org/10.1111/j.1600-051X.2009.01408.x
16. Akinkugbe AA, Saraiya VM, Preisser JS, Offenbacher S, Beck JD. Bias in estimating the cross-sectional smoking, alcohol, obesity and diabetes associations with moderate-severe periodontitis in the Atherosclerosis Risk in Communities study: comparison of full versus partial-mouth estimates. J Clin Periodontol. 2015 Jul;42(7):609-21. https://doi.org/10.1111/jcpe.12425
17. Research, Science and Therapy Committee. Position Paper: Diagnosis of Periodontal Diseases. J Periodontol. 2003 Aug;74(8):1237-1247. https://doi.org/10.1902/jop.2003.74.8.1237
18. Kingman A, Albandar JM. Methodological aspects of epidemiological studies of periodontal diseases. Periodontol 2000. 2002;29:11-30. https://doi.org/10.1034/j.1600-0757.2002.290102.x
19. Leroy R, Eaton KA, Savage A. Methodological issues in epidemiological studies of periodontitis--how can it be improved? BMC Oral Health. 2010 Apr 21;10:8. https://doi.org/10.1186/1472-6831-10-8
19. Leroy R, Eaton KA, Savage A. Methodological issues in epidemiological studies of periodontitis--how can it be improved? BMC Oral Health. 2010 Apr 21;10:8. https://doi.org/10.1186/1472-6831-10-8
20. Romano F, Perotto S, Castiglione A, Aimetti M. Prevalence of periodontitis: misclassification, under-recognition or over-diagnosis using partial and full-mouth periodontal examination protocols. Acta Odontol Scand. 2019

Apr;77(3):189-196. https://doi.org/10.1080/00016357.2018.1535136
21. Susin C, Kingman A, Albandar JM. Effect of partial recording protocols on estimates of prevalence of periodontal disease. J Periodontol. 2005 Feb;76(2):262-7. https://doi.org/10.1902/jop.2005.76.2.262
22. Tran DT, Gay I, Du XL, Fu Y et al. Assessment of partial-mouth periodontal examination protocols for periodontitis surveillance. J Clin Periodontol. 2014 Sep;41(9):846-52. https://doi.org/10.1111/jcpe.12285
23. World Health Organization. Oral health surveys: basic methods - 4th edition. 1997. https://apps.who.int/iris/handle/10665/41905
24. Roncalli AG, Silva NN, Nascimento AC, Freitas CH et al. Aspectos metodológicos do Projeto SBBrasil 2010 de interesse para inquéritos nacionais de saúde [Relevant methodological issues from the SBBrasil 2010 Project for national health surveys]. Cad Saude Publica. 2012;28 Suppl:s40-57. https://doi.org/10.1590/S0102-311X2012001300006
25. World Health Organization. Oral health surveys: basic methods - 5th edition. 2013. https://www.who.int/publications-detail-redirect/9789241548649
26. Ministerio de Salud Pública. 2ª Encuesta nacional de factores de riesgo de enfermedades crónicas no trasmisibles. Montevideo: MSP; 2013 http://www.msp.gub.uy
27. Unger T, Borghi C, Charchar F, Khan NA, Poulter NR, Prabhakaran D, Ramirez A, Schlaich M, Stergiou GS, Tomaszewski M, Wainford RD, Williams B, Schutte AE. 2020 International Society of Hypertension Global Hypertension Practice Guidelines. Hypertension. 2020 Jun;75(6):1334-1357. https://doi.org/10.1161/HYPERTENSIONAHA.120.15026
28. R Core Team. R: A language and environment for statistical computing. [Internet]. R Foundation for Statistical Computing, Vienna, Austria. 2020. https://www.r-project.org/
29. Peres MA, Peres KG, Cascaes AM, Correa MB et al. Validity of partial protocols to assess the prevalence of periodontal outcomes and associated sociodemographic and behavior factors in adolescents and young adults. J Periodontol. 2012 Mar;83(3):369-78. https://doi.org/10.1902/jop.2011.110250
30. Klinge B, Norlund A. A socio-economic perspective on periodontal diseases: a systematic review. J Clin Periodontol. 2005;32 Suppl 6:314-25. https://doi.org/10.1111/j.1600-051X.2005.00801.x
31. Enwonwu CO, Salako N. The periodontal disease-systemic health-infectious disease axis in developing countries. Periodontol 2000. 2012 Oct;60(1):64-77. https://doi.org/10.1111/j.1600-0757.2012.00447.x
32. Fi C, Wo W. Periodontal disease and systemic diseases: an overview on recent progresses. J Biol Regul Homeost Agents. 2021;35(1):1–9.
33. Lorenzo-Erro SM, Skapino E, Musto M, Olmos P et al. Oral Health and Non communicable Diseases in patients of a higher education institution, Montevideo, Uruguay 2016. Part 1. Odontostomatology. 2020 Dec 12;22(36):55–64. https://doi.org/10.22592/ode2020n36a7
34. Niskanen MC, Mattila PT, Niinimaa AO, Vehkalahti MM et al. Behavioural and socioeconomic factors associated with the simultaneous occurrence of periodontal disease and dental caries. Acta Odontol Scand. 2020 Apr;78(3):196-202. https://doi.org/10.1080/00016357.2019.1679389
35. Sun HY, Jiang H, Du MQ, Wang X, Feng XP, Hu Y, Lin HC, Wang B, Si Y, Wang CX, Zheng SG, Liu XN, Rong WS, Wan WJ, Tai BJ. The Prevalence and Associated Factors of Periodontal Disease among 35 to 44-year-old Chinese Adults in the 4th National Oral Health Survey. Chin J Dent Res. 2018;21(4):241-247. https://doi.org/10.3290/j.cjdr.a41082
36. Wang J, Lv J, Wang W, Jiang X. Alcohol consumption and risk of periodontitis: a meta-analysis. J Clin Periodontol. 2016 Jul;43(7):572-83. https://doi.org/10.1111/jcpe.12556
37. Pulikkotil SJ, Nath S; Muthukumaraswamy; Dharamarajan L, Jing KT, Vaithilingam RD. Alcohol consumption is associated with periodontitis. A systematic review and meta-analysis of observational studies. Community Dent Health. 2020 Feb 27;37(1):12-21. doi: 10.1922/CDH_4569Pulikkotil10. https://doi.org/10.1922/CDH_4569Pulikkotil10.
38. Jobin K, Stumpf NE, Schwab S, Eichler M et al. A high-salt diet compromises antibacterial neutrophil responses through hormonal perturbation. Sci Transl Med. 2020 Mar 25;12(536):eaay3850. https://doi.org/10.1126/scitranslmed.aay3850
39. Iwasaki M, Moynihan P, Manz MC, Taylor GW, Yoshihara A, Muramatsu K, Watanabe R, Miyazaki H. Dietary antioxidants and periodontal disease in community-based older Japanese: a 2-year follow-up study. Public Health Nutr. 2013 Feb;16(2):330-8. https://doi.org/10.1017/S1368980012002637
40. Kondo K, Ishikado A, Morino K, Nishio Y et al. A high-fiber, low-fat diet improves periodontal disease markers in high-risk subjects: a pilot study. Nutr Res. 2014 Jun;34(6):491-8. https://doi.org/10.1016/j.nutres.2014.06.001
41. Benigeri M, Brodeur JM, Payette M, Charbonneau A et al. Community periodontal index of treatment needs and prevalence of periodontal conditions. J Clin Periodontol. 2000 May;27(5):308-12. https://doi.org/10.1034/j.1600-051x.2000.027005308.x

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Universal adhesives applied to deep dentin with different bonding treatments

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ABSTRACT

Eighth-generation adhesives may be applied with total etch, selective-etch or self-conditioning, and serve as primers for non-dental substrates. ***Aim:*** *To determine the bonding characteristics of universal adhesives applied to the deep pulp wall with different strategies, by means of shear bond strength and laser microscopy.* ***Materials and Method:*** *Cavities 4 mm deep and maximum width were carved in 36 extracted molars. Nine groups were formed according to dental substrate treatment and adhesives, as follows:* **Total-etch:** *group 1-Monobond 7 self-etch, group 2-One coat 7 universal, and group 3-Single bond universal;* **Adamantine etch:** *group 4-Monobond 7 self-etch, group 5-One coat 7 universal, and group 6-Single bond universal;* **Self-conditioning:** *group 7-Monobond 7 self-etch, group 8-One coat 7 universal, and group 9-Single bond universal. Molars were filled following the manufacturer's instructions. Three specimens per group (27 altogether) were used to determine shear bond strength using a universal testing machine, while layer thicknesses were measured on the remaining specimens using microscope images and Olympus LEXT 3D Software. Analysis of variance was used to compare data.* ***Results:*** *Mean (standard deviation) bond strength in megapascals (MPa) was: group 1: 7.06±3.01; group 2: 10.74±4.36; group 3: 8.20±3.92; group 4: 7.41±2.23; group 5: 6.84±1.50; group 6: 5.86±2.10; group 7: 5.83±1.94; group 8: 7.14±2.37; group 9: 8.06±3.51. Bond strength was higher (p=0.049) for total-etch (8.61±3.96) than for selective etch (6.71±1.98) and self-conditioning (6.91±2.68). No significant difference was found among the three adhesives (p=0.205). Adhesive layer in micrometers (µm) was total-etch 8.71±4.93, selective etch 5.49±1.70 and self-conditioning 6.27±3.01, with no significant difference.* ***Conclusions:*** *There were significant differences among bonding strategies, with the highest values for total-etch. No significant difference was observed between self-conditioning and selective etch. No significant difference was found among the adhesives, which all behaved similarly. The greatest adhesive layer thicknesses were recorded in the total-etch group, with no significant difference among the various adhesive approaches.*

Keywords: *dentin bonding agents - microscopy confocal*

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Adhesivos universales aplicados a dentina profunda con diferentes tratamientos de unión

RESUMEN

Los adhesivos universales de octava generación pueden ser aplicados con diferentes estrategias de unión: grabado total, grabado selectivo o autoacondicionamiento. Además, imprimen sustratos no dentales. ***Objetivo:*** *Determinar las caracteristicas de unión de adhesivos universales con diferentes estrategias en pared pulpar profunda mediante resistencia adhesiva al corte y microscopía laser.* ***Materiales y Método:*** *En 36 molares se tallaron cavidades de 4 mm de profundidad y ancho máximo. Se dividieron en 9 grupos según tratamientos y adhesivos.* **Grabado total:** *grupo 1-Monobond 7 self-etching, grupo 2-One coat 7 universal y grupo 3-Single bond universal;* **Grabado selectivo:** *grupo 4-Monobond 7 self-etching; grupo 5-One coat 7 universal y grupo 6-Single bond universal y* **Autoacondicionamiento:** *grupo 7-Monobond 7 self-etching; grupo 8-One coat 7 universal y grupo 9-Single bond universal. Las obturaciones se realizaron siguiendo las instrucciones del fabricante. La resistencia adhesiva al corte se determinó utilizando una máquina de ensayo universal sobre 27 especímenes mientras que los restantes fueron empleados para evaluar los espesores de la capa generado sobre imágenes obtenidas con microscopía y con el software Olympus LEXT 3D. Se ultilizó análisis de varianza.* ***Resultados:*** *Resistencia adhesiva en megapascal (MPa) media (desviación estándar): grupo 1: 7,06±3,01; grupo 2: 10,74±4,36; grupo 3: 8,20±3,92; grupo 4: 7,41±2,23; grupo 5: 6,84±1,50; grupo 6: 5,86±2,10; grupo 7: 5,83±1,94; grupo 8: 7,14±2,37; grupo 9: 8,06±3,51. Grabado total (8,61±3,96) registró los valores mayores (p=0,049) en comparación a grabado selectivo (6,71±1,98) y autoacondicionamiento (6,91±2,68). Los adhesivos no tuvieron diferencias significativas (p=0,205). Capa adhesiva en µm: Grabado total (8,71±4,93); grabado selectivo (5,49±1,70) y autoacondicionamiento (6,27±3,01) sin diferencias significativas (p=0,073).* ***Conclusiones:*** *Las estrategias de unión mostraron diferencias significativas; los valores más altos se obtuvieron con grabado total y entre autoacondicionamiento y grabado selectivo no hubo significancia. Los adhesivos evidenciaron comportamientos similares sin registrar diferencias significativas. Los mayores espesores de capa fueron con grabado total sin diferencias significativas entre las técnicas.*

Palabras clave: *adhesivos dentales - microscopía confocal*

INTRODUCTION

Universal dental adhesives were introduced as versatile multifunctional systems with fewer application steps, compatible with all treatment modalities for mineralized dental tissues[1]. Universal adhesives, also known as multimode or multipurpose adhesives, have been used in clinical practice since 2011. They can be applied to enamel and dentin as self-conditioning adhesives, and to enamel as etch-rinse adhesives, a technique known as "selective enamel etching". The acid monomers in their composition act as primers for non-dental substrates (alloys and polycrystalline ceramics), making universal adhesives suitable for application with different bonding strategies[2,3]. These adhesives have been recently called eighth-generation adhesives, based on the historical perspective of dental adhesives[4].

Universal adhesives contain mixed monomers with slight to moderate acidity in reduced concentrations, conventional dimethacrylate cross-linkers, non-acid emulsifying monomers, catalyzer for light and dual curing and an adequate selection of solvents to improve monomer spreading and substrate infiltration capacity[2,5].

An adhesive that may be used in different procedures enables the dentist to choose the technique that best suits the clinical case, thereby optimizing the final restorative result. For instance, when a restoration requires a strong bond to the enamel, or in case of sclerotic dentin, it may be advisable to perform previous etching. The etching stage can be graded according to the time for which the phosphoric acid gel is applied before rinsing. On the other hand, it may be preferable to use the self-conditioning technique when dealing with difficult surgical access, limited time, or non-collaborative very young patients[6].

Even though it has been scientifically documented that the etching technique using phosphoric acid enhances one-step self-conditioning bonding to enamel, it is advisable to take more care with the etching procedure using additional phosphoric acid on dentin[7]. This is probably the main reason why most universal adhesive contain 10-methacryloxydecyl dihydrogen phosphate (10-MDP), a functional group of phosphoric acid, as their main adhesive monomer which produces a limited decalcification effect on the dentin surface. This procedure is considered the most reliable treatment for dentin, since the universal adhesives containing MDP have slight acidity and the capacity to interact chemically with hydroxyapatite crystals and enable stable salt formation of calcium phosphate and calcium carboxylate insoluble in water[8,9].

This new philosophy of versatile bonding encourages the use of the simplest option for each strategy, i.e., one-step self-conditioning or two-step etch and rinse, in order to bond direct or indirect restorations to enamel and dentin[10]. The universal etch and rinse (E&R) bonding mode involves a phosphoric acid etching step followed by a thorough water rinsing phase prior to application of a primer/adhesive resin combination. Monomers diffuse into the micro-etch pits created on the enamel to form microtags and macrotags, and into the exposed collagen fibril network at dentin to form a 3-5 µm hybrid layer. While the E&R bonding mode is undoubtedly the best bonding strategy to enamel, the resultant thick and HAp-free hybrid layer formed on the dentin is highly sensitive to degradation over time. The universal self-etch (SE) bonding mode involves the use of monomers with an acidic functional group that in principle simultaneously etches and infiltrates dentin down to a depth of about 1 µm. In general, the SE bonding mode underperforms the E&R bonding mode on enamel, by which enamel remains to be selectively etched with phosphoric acid. SE bonding nevertheless possesses chemical bonding potential as an additional benefit to achieve durable bonding[4]. However, universal bonding systems may show the deficiencies of their predecessors, i.e., one-step systems[11], so their bonding performance to dentin should be evaluated using different variables that modify in-vitro bond strength[12]. In this context, the purpose of the present study was to use shear bond strength and laser microscopy tests to determine the bonding characteristics of universal adhesives using total enamel and dentin etching treatments, adamantine selective etching and self-conditioning on the pulp wall of deep cavity preparations.

MATERIALS AND METHOD

This study was approved by the Academic Committee for Health Research (*Comité Académico de Investigaciones en Salud*) of the School of Dentistry, National University of Cordoba (CIEIS-ODO-CASI 48 I).

Cavity preparation

Single occlusal cavities 4 mm deep and maximum

width were carved in 36 third molars which had been extracted for orthodontic reasons. Cavity depth standardization was checked using a periodontal millimeter probe, CP-11, Hu-Friedy, Illinois, United States. The molars were divided into nine groups according to dentin substrate treatment (total etching, selective adamantine etching or self-conditioning), and the universal adhesive applied. **Group 1:** Total-etch+Monobond 7 self-etch adhesive (Densell, Buenos Aires, Argentina). **Group 2:** Total-etch+One coat 7 universal adhesive (Coltene, Altstätten, Switzerland). **Group 3:** Total-etch+Single bond universal adhesive (3M ESPE, Neuss, Germany). **Group 4:** Adamantine etching+Monobond 7 self-etch adhesive (Densell, Buenos Aires, Argentina). **Group 5:** Adamantine etching+One coat 7 universal adhesive (Coltene, Altstätten, Switzerland). **Group 6:** Adamantine etching+Single bond universal adhesive (3M ESPE, Neuss, Germany). **Group 7:** Monobond 7 self-etch adhesive (Densell, Buenos Aires, Argentina). **Group 8:** One coat 7 universal adhesive (Coltene, Altstätten, Switzerland). **Group 9:** Single bond universal adhesive (3M ESPE, Neuss, Germany). In groups 1, 2 and 3, total-etch was performed on enamel and dentin using 35% phosphoric acid (Densell, Buenos Aires, Argentina) for 10 s, followed by spray washing and drying for 5 s. Finally, universal bonding was applied according to the manufacturer's instructions. In groups 4, 5 and 6, enamel was etched using 35% phosphoric acid (Densell, Buenos Aires, Argentina) for 10 s. In groups 7, 8 and 9, universal adhesives were applied directly on-to the enamel and dentin as indicated by the manufacturer.

All the cavities were filled with submicron-hybrid composite resin Brilliant EverGlow (Coltene, Altstätten, Switzerland), by means of oblique incremental technique, and each layer was polymerized for 20 s with a LED unit (Optilux LED, Coltene, Altstätten, Switzerland).

Shear bond strength

Three specimens from each group (27 altogether) were first sectioned longitudinally in lingual-buccal direction into sections 2.5 mm and 3 mm thick on average; then transversally, using a hard tissue microtome Isomet (Buehler Co., Illinois, United States) at 300 rpm and 50 grams pressure, under continuous water cooling. Two test specimens averaging 2.6 mm wide by 2.6 mm high were thus formed, which included dentin pulp wall, universal adhesive system and composite resin. Thus, each group consisted of six test specimens (n=6). These samples were stored at 37 ºC for 24 h in a 100% humidity atmosphere.

Shear bond strength tests were performed using a universal testing machine with a crosshead speed of 0.5 mm per minute. The sections were fixed with a dental press and placed on the plate of the device such that the composite resin/dentin pulp wall union remained next to the edge. The beveled edge of the shear was placed at 0.5 mm from the material/substrate joint for cutting. A standard 0.5 mm thick layer was used to corroborate the separation. Analysis of variance was applied to determine the efficacy of the substrate treatment with each universal adhesive used.

Laser confocal microscopy study

One specimen from each group (9 altogether) was sectioned longitudinally using a hard tissue microtome Isomet (Buehler Co., Illinois, United States) at 300 rpm and 50 g pressure under continuous water cooling, to obtain test specimens 1.5 mm to 2 mm thick for each group (n=3 for each group). The sections were polished using a metallographic polisher (Praxis, Buenos Aires, Argentina) with tapered granulometry disks and felt cloths, washed with ultrasound and stored in a stove at 37 ºC for 24 h in a 100% humidity atmosphere. The sections were observed with laser confocal microscopy, Lext 3D Measuring Laser Microscope OLS4000 (Olympus, Japan). The thicknesses of the adhesive layer on the deep pulp walls were measured using the same software. Ten measurements were performed by microphotography at regular intervals on 50 µm long paths (Fig. 1). The data were analyzed using analysis of variance. Statistical significance was set at 0.05 for all tests.

RESULTS

Shear bond strength

Considering each adhesive with its respective treatment, shear bond strength was significantly higher (p=0.024) for One coat 7 universal using total-etch procedure than for any of the other three treatments performed on the dental substrate. Monobond 7 self-etch had the highest values with selective enamel etching, with no significant difference (p=0.384). Single bond universal with

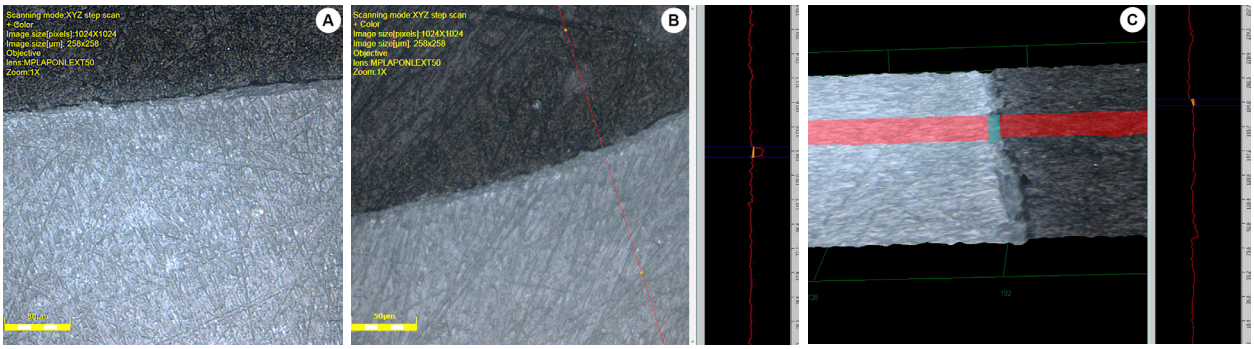

Fig. 1: *B and C show profile sections that were measured.* **A)** *Group 1: Simultaneous etching technique of enamel and dentin and application of Universal Adhesive Monobond 7 self-etch. Mean layer thickness between the pulp wall and the restoration material was 8.71±4.93 µm.* **B)** *Group 5: Application of One Coat Universal Adhesive by means of selective enamel etching. Mean layer thickness was 5.49±1.70 µm, and* **C)** *Application of Single Bond Universal Adhesive through self-etch mode. Mean layer thickness was 6.27±3.01 µm.*

total-etch protocol had higher values than the other treatments, with no statistically significant difference (p=0.299) (Fig. 2).

In general, bond strength for the total-etch treatment (8.61±3.96) was significantly greater than for the other treatments: selective enamel etching (6.71±1.98) and self-conditioning (6.91±2.68); p=0.049 (Fig. 3).

There was no statistically significant difference (p=0.205) among the different universal adhesives used. One coat 7 universal (8.33±3.45) mean values were higher than those of the other universal adhesives, Single bond universal (7.38±3.33) and Monobond 7 self-etch (6.75±2.47) (Fig. 4).

Laser confocal microscopy analysis: Adhesive layer thickness

Adhesive layer thicknesses were greatest for the total-etch treatment group, followed by self-conditioning, and lowest for the selective enamel etching treatment group. There was no significant difference among the different adhesive techniques (Fig. 5).

DISCUSSION

The present study found that certain in-vitro conditions such as deep pulp wall and different universal adhesive bonding approaches determined low bonding values expressed in MPa in the shear bond strength tests when compared to those obtained,

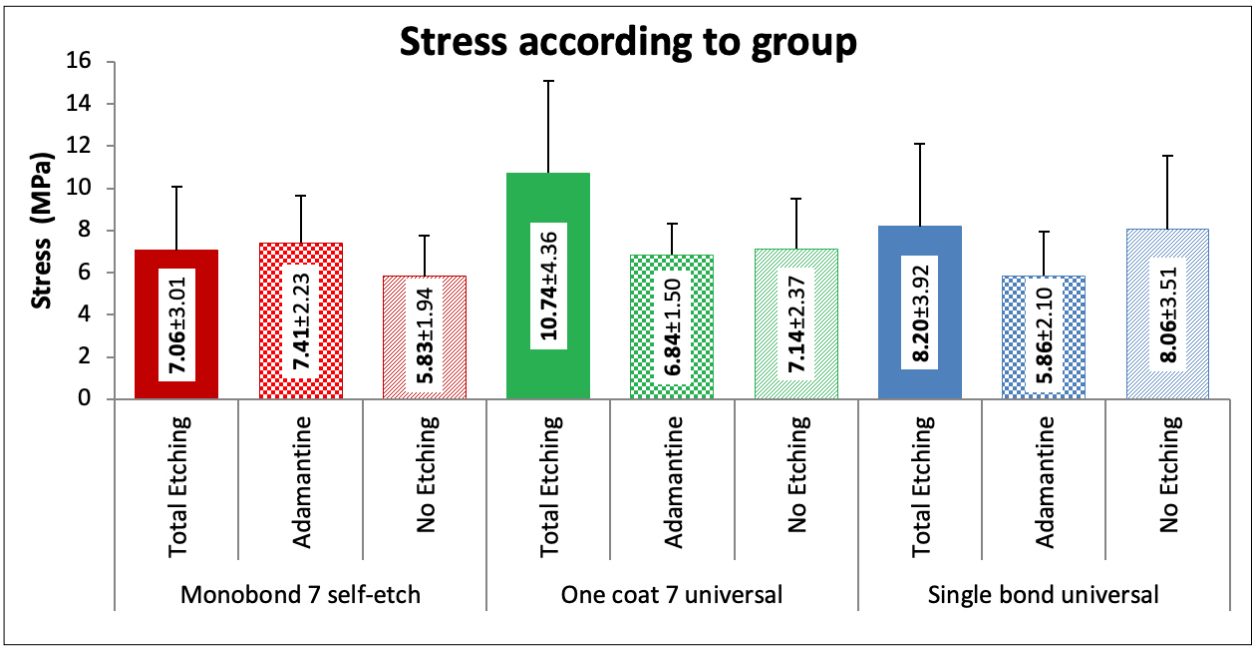

Fig. 2: *Shear stress according to adhesive and technique used. Mean and standard deviation values expressed in MPa.*

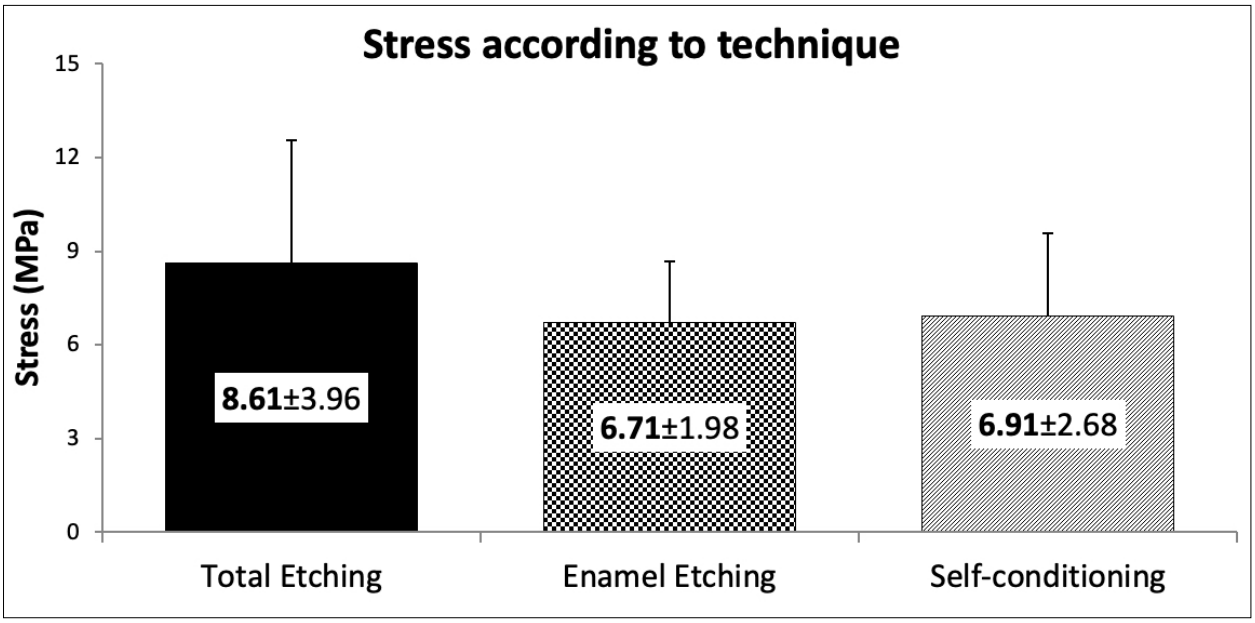

Fig. 3: *Shear stress according to technique. Mean and standard deviation values expressed in MPa.*

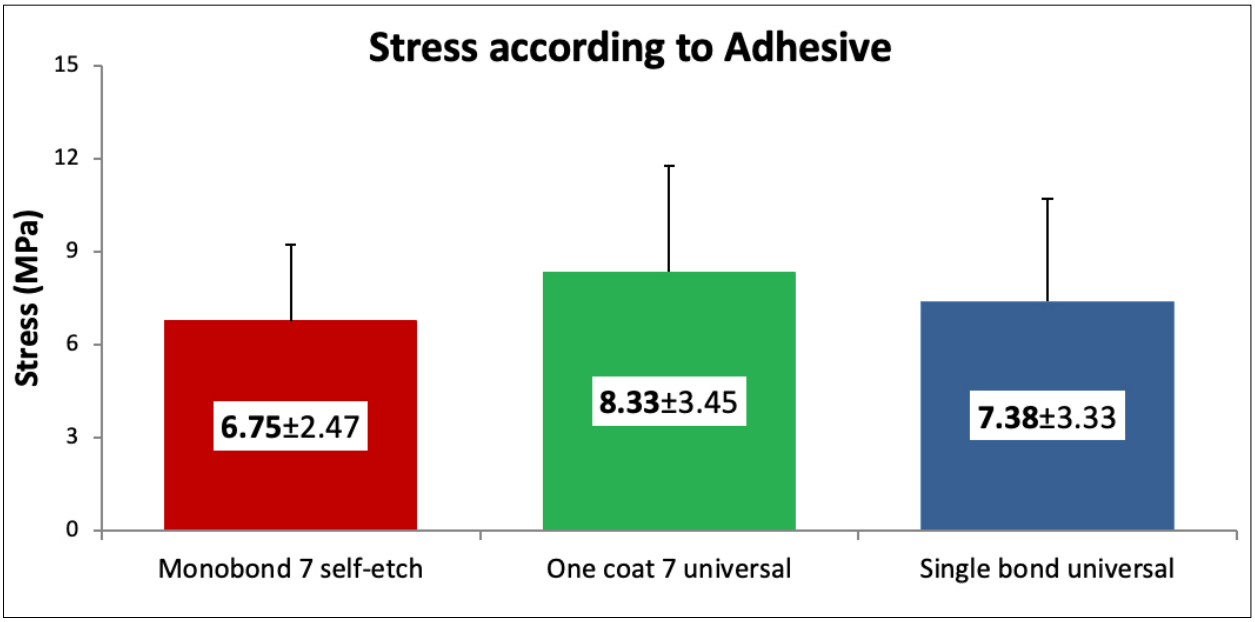

Fig. 4: *Shear stress according to universal adhesive. Mean and standard deviation values expressed in MPa.*

for example, for lateral, median or superficial walls[13]. This means that cavity depth and wall type affect the results of the laboratory tests, regardless of the bonding approach chosen. We agree that it is more difficult to achieve bonding to the dentin closer to the pulp chamber roof than to the superficial dentin[4]. Adhesion to dentin remains a challenge, mainly because dentin has a more organic composition than enamel, and its wet, organic nature makes bonding difficult[13]. When dentin is etched, the acid demineralizes the intertubular dentin, resulting in the exposure of the superficial collagen network[14]. The network is infiltrated with the adhesive resin, leading to the formation of a hybrid layer which is responsible for the bond between the resin and the dentinal tissues[15]. To ensure ideal bonding conditions, the dentin demineralized with acid must be kept wet to prevent the collagen fibrils from collapsing, though not too wet, because over-wetting prevents resin impregnation of the collagen fibrils[16]. Smear layer removal with single-step etching before the dentin adhesive is applied (etch-rinse technique), or its modification with a self-etch monomer (self-conditioning technique) is crucial to form a hybrid

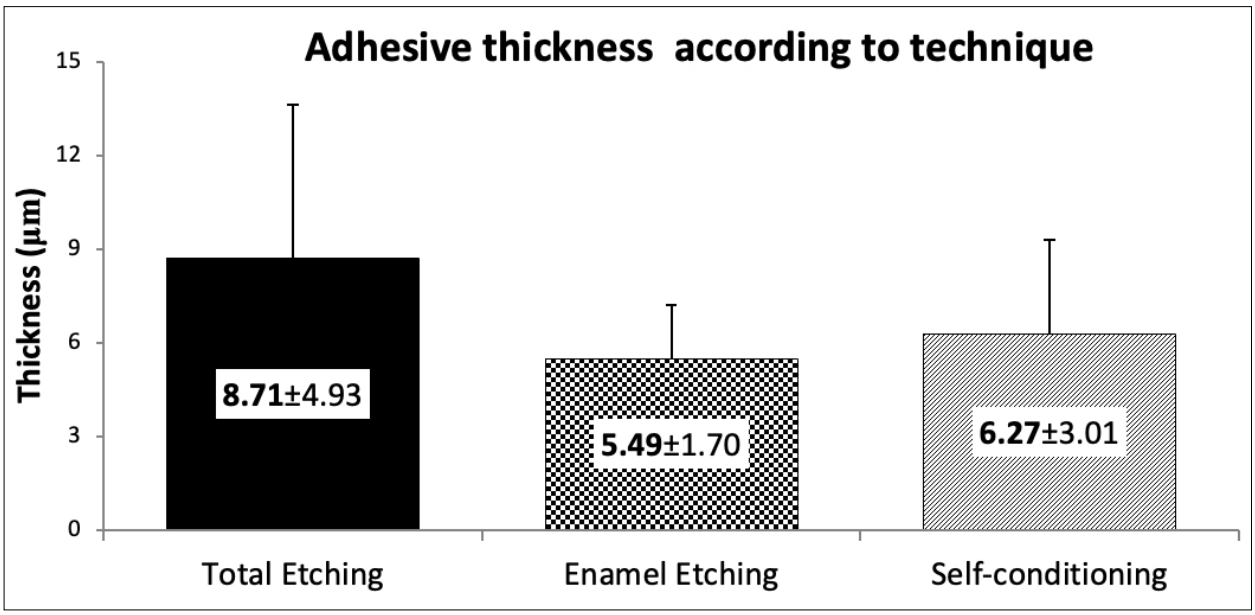

Fig. 5: *Adhesive layer thickness according to technique. Mean and standard deviation values expressed in µm.*

layer in order to ensure an effective bond between the adhesive resin and the dentin[17].

The present study found higher values for total-etch treatments in deep dentin than for the other two methods (selective etching and self-conditioning). However, during the total conditioning step, phosphoric acid eliminates the smear layer while demineralizing the dentin to a depth of 3 to 5 mm, thus exposing a collagen fibril scaffold deprived of hydroxyapatite[18,19]. The highly mineralized peritubular dentin dissolves almost completely and the dentin tubules widen. However, the literature reports that no statistically significant difference was recorded in the dentin regarding bonding efficacy when self-etch or etch-rinse approaches were used[2]. It has been suggested that the bonding techniques requiring smear layer removal are associated with greater postoperative sensitivity than systems that leave the smear layer in situ[20]. Even though it has been demonstrated that the bonding procedure may cause transitory pulp inflammation, especially in the deep cavities, it is likely that continuous bacterial irritation due to microgaps and microfiltration may cause damage to the pulp and postoperative pain[21].

Shear bond strength is a valid versatile method for assessing bonding effectiveness to dental substrates in laboratory tests[22]. The formation of smear layer in vitro has also been shown in laboratory tests. Chowdhury et al. assessed the effects of the smear layers formed by abrasives having similar coarseness to the fine-grit diamond stone. They established a model for dentin bonding tests, yielding clinically relevant significant results. They also performed micro-tensile bond strength tests (µTBS) of currently available universal adhesives and of a two-step self-etch adhesive. The presence of smear layer formed under these conditions had no significant effect on the resin-dentin bond strength of the adhesives tested. Moreover, the performance of the bond of the universal adhesives to the dentin may be improved duplicating its application time. The elimination or modification of the smear layers covering the dentin is critical to allow penetration of the adhesive molecules and to ensure a strong bond between the resin and the dentin[23]. Universal adhesives benefit from phosphoric acid etching because bond strengths increase, mainly on the enamel surface. The authors compared in vitro shear strength of four universal adhesives on enamel and dentin with and without additional phosphoric acid etching, finding mean bond strength values to enamel ranging from 13.4 and 21.9 MPa in the self-etch mode. When the etch-rinse protocol was used, the mean bond strength was over 30 MPa. Regarding the dentin, the significant differences in the self-etch mode depended on each adhesive used[24]. Stape et al. claim that selective dentine etching for 3 s improved the interaction depth of the tested universal adhesive without overexposing the demineralized collagen or reducing Ca availability

at the bonded interface. Nevertheless, the universal adhesives used in the self-etch mode produce superior long-term dentin bonding compared to the etch and rinse mode. Selective etching for 3 s with conventionally used H_3PO_4 improves dentin bonding effectiveness; nonetheless, longer etching times should be strictly avoided[25]. With regard to the self-conditioning protocol, Tsujimoto et al. compared universal adhesives in self-conditioning mode and two-step self-conditioning adhesives by means of initial shear bond strength tests and shear-fatigue strength test, at the level of the resin composite/adhesive bond to dentin. Their results encourage the continued use of the two-step self-etch adhesive over some universal adhesives but suggest that changes to the composition of universal adhesives may lead to dentin bond fatigue durability similar to that of two-step self-etch adhesives[26]. Daneshkazemi et al. reported mean values in MPa between 35.74 and 18.09 in superficial dentin in micro-tensile with two adhesive protocols, self-etch and etch-rinse. Universal adhesives had the highest adhesive values considered as independent etching[27]. Lezaja Zebic et al. conducted microtensile bond strength tests of universal adhesives applied to dentin following total etch or self-etch protocols, direct or indirect water storing, using pulpal pressure simulation. Adhesives were applied to class 1 cavities and to mid-coronal dentin. Results obtained following the self-etch protocol were more stable in the long term than with total-etch protocol. Simulated pulpal pressure and Class 1 preparation may be recommended for adhesive strength tests. The values obtained were in the range of 19-42 MPa initially and 16-36 MPa after 6 months storage[28].

Concerning the effect of the chemical composition of the adhesives on the bond strength with dental substrates, Papadogiannis et al. performed bond strength tests of universal adhesives based on the adhesive monomer 10-MDP. Adhesive monomer, the inclusion of different comonomers (reticulating or bond promotors) catalyzers and solvents led to great variations in the properties of the adhesive film, thus affecting its reactivity with dentin and later its bond strength. By using infrared reflectance microscopy, the authors confirmed that the dentin surfaces treated with universal adhesives did not have a smear layer. Moreover, microscopic images exhibited gaps and porosities[29].

By using an electron microscope, Zhang et al. observed collagen degradation not seen on hybrid layers created by adhesives containing 10-MDP with the etch-rinse mode, which produced collagen fibers that were partially degraded with intact periphery[30]. In line with this, the results reported by Zecin-Deren et al. are attributed to the fact that the adhesive used contains 10-MDP as adhesive monomer in its chemical composition. In bond strength tests, these authors found higher mean values with Single Bond Universal than with other adhesives used in their study[31]. This phosphoric acid functional group also contains a polymerizable methacrylate group responsible for the curing potential, and a group of 10-carbon chain to separate the other two active groups[32].

The carbon separator affects the monomer flexibility, solubility, moisturizing, and hydrophilic-hydrophobic balance[33]. In order to further improve bond strength to dentin of Single Bond Universal, it may be advisable to apply it two or three times and to polymerize it after the final application[31].

Taking dentin depth and conditioning mode as variables, Yousry et al. reported that using etch-rinse adhesives, the shear bond strength values in superficial dentin were significantly higher than in deep dentin. Unlike the results obtained with the self-etch-systems, the performance in both dentins was similar. The authors concluded that bond strength to dentin depends on both the adhesive and the substrate. Contemporary adhesive systems may produce variable bond results to superficial and deep dentin owing to variations in their composition rather than to their application technique[34].

Similarly, Yoshihara et al. concluded that the 10-MDP monomer in high purity is essential to achieve long-lasting bonding, excellent hybridization with 10-MDP-Ca salts, and nanolayering. They suggested that the highest bond effectivity of 10-MDP-based adhesives reported are not only attributed to a stronger 10-MDP chemical bond, but also to higher etching potential[35].

Rosa et al. conducted a systematic review of 10 articles to determine whether etch-rinse mode or self-etch mode is the better protocol for enamel and dentin bonding by universal adhesives. The in vitro studies analyzed the bond strength of universal adhesives to dentine and/or enamel through the etch-rinse and self-etching strategies. They concluded that the bond strength to enamel of universal adhesives is enhanced by previous etching with phosphoric

acid. However, this effect was not evident on dentin with the use of mild universal adhesives with etch-rinse differences strategy. No statistically significant difference was found between etch-rinse and self-etch for mild universal adhesives[36].

Regarding the different protocols in which the universal adhesives can be used and the way these modes may affect and modify dentin wetness, Kumagai et al. claimed that universal adhesives may be applied with either self-etch or etch-rinse modes. Nevertheless, universal adhesives should be strictly applied to wet dentin for bonding in the etch-rinse mode. They observed a well-formed hybrid layer on wet dentin, in contrast to defects, pores and reduced hybridization thickness when the adhesives were applied to over-dried dentin[37].

Choi et al. analyzed wetness on the dentin surface as a factor affecting the micro-tensile bond strength of universal adhesives. They suggest that the wetness of the dentin surface should be carefully controlled with special consideration for the application of universal adhesives[38]. Sugimura et al. published that some universal adhesives, with the addition of specific components and water content optimization, can achieve stable bonds irrespective of surface wetness, though they agreed that the moisture of the dentin surface is an important factor for universal adhesive bonding in the etch-rinse mode. In addition, dentin surface wetness did not influence the thickness of the adhesive layer or hybrid layer of the dentine-resin interfaces[39].

In the present study, laser microscopy measurements showed that the adhesive layer was thicker in the total-etch treatment group (8.71 μm), and thinner in both the self-etch approach (6.27 μm) and selective etching procedure in enamel (5.49 μm). There were no significant differences among the adhesive techniques proposed. These values agree with a publication which reports that the etch-rinse approach increases hybridization thickness (4 to 6 μm)[4]. Chen et al. observed hybrid layers of universal adhesives ~5 μm and <0.5 μm thick in the etch-rinse mode and self-conditioning mode[5]. By using electron microscopy, Takamizawa et al. observed similar adhesive layer thicknesses for single-step universal adhesives and self-conditioning adhesives[40].

Universal adhesives reflect manufacturers' efforts to provide versatility in product design by adapting a single bottle self-etch adhesive to other application modes, without compromising its bonding effectivity[37]. These adhesives may be applied in simplified clinical steps, are less technique-sensitive, require shorter application times, and cause less postoperative sensitivity. The present study showed that the highest values were obtained with the total etching mode. Moreover, if this protocol were used in deep dentin, universal adhesives would not fulfill the premise that they cause little or no postoperative sensitivity, a highly relevant clinical factor in selecting the best technique for dentinal substrate treatment.

In conclusion, in the present study, shear bond strength using various protocols for activation of deep dentine substrates differed significantly among the three conditioning procedures. The total etching treatment or total removal of the smear layer yielded the highest bond strength values analyzed in the deep pulp wall. The bonding performance of universal adhesives applied with self-conditioning approach was similar to that of selective enamel etching protocols. In general, bond strength values did not differ significantly among self-etch Monobond 7, One coat 7 universal and Single bond universal. The bond layer was thickest in the total etching treatment, without significant differences among the three bonding techniques applied. Regardless of the different results obtained in laboratory tests, it is the dentist who should decide on the most appropriate mode for the various clinical situations, especially when the deep dentin wall is involved.

DECLARATION OF CONFLICTING INTERESTS

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REFERENCES

1. Nagarkar S, Theis-Mahon N, Perdigão J. Universal dental adhesives: Current status, laboratory testing, and clinical performance. J Biomed Mater Res B Appl Biomater. 2019;107:2121-2131. https://doi.org/10.1002/jbm.b.34305
2. Hanabusa M, Mine A, Kuboki T, Momoi Y et al. Bonding effectiveness of a new "multi-mode" adhesive to enamel and dentine. J Dent. 2012;40(6):475-484. https://doi.org/10.12691/ijdsr-6-5-3

3. Perdigão J, Sezinando A, Monteiro PC. Laboratory bonding ability of a multi-purpose dentin adhesive. Am J Dent. 2012; 25:153-158.
4. Van Meerbeek B, Yoshihara K, Van Landuyt K, Yoshida Y et al. From Buonocore's pioneering acid-etch technique to self-adhering restoratives. A status perspective of rapidly advancing dental adhesive technology. J Adhes Dent. 2020;22(1):7-34. https://doi.org/10.3290/j.jad.a43994
5. Chen C, Niu LN, Xie H, Zhang ZY et al. Bonding of universal adhesives to dentine-Old wine in new bottles? J Dent. 2015;43(5):525-536. https://doi.org/10.1016/j.jdent.2015.03.004
6. Sofan E, Sofan A, Palaia G, Tenore G et al. Classification review of dental adhesive systems: from the IV generation to the universal type. Ann Stomatol (Roma). 2017;8(1):1-17.
7. Van Meerbeek B, Yoshihara K, Yoshida Y, Mine A et al. State of the art of self-etch adhesives. Dent Mater. 2011;27:17-28. https://doi10.1016/J.dental.2010.10.023
8. Wang R, Shi Y, Pan Y, Cui Y et al. Adhesive interfacial characteristics and the related bonding performance of four self-etching adhesives with different functional monomers applied to dentin. J Dent. 2017;62:72-80. https://doi.org/10.1016/j.jdent.2017.05.010
9. Carrilho E, Cardoso M, Marques Ferreira M, Marto CM et al. 10-MDP based dental adhesives: Adhesive interface characterization and adhesive stability-A systematic review. Materials. 2019;12(5):790. https://doi.org/10.3390/ma12050790
10. Muñoz MA, Luque I, Hass V, Reis A et al. Immediate bonding properties of universal adhesives to dentine. J Dent. 2013;41(5):404-411. https://doi.org/10.1016/j.jdent.2013.03.001
11. Zhang ZY, Tian FC, Niu LN, Ochala K et al. Defying ageing: An expectation for dentine bonding with universal adhesives? J Dent. 2016;45:43-52. https://doi.org/10.1016/j.jdent.2015.11.008
12. Miyazaki K, Tsubota K, Takamizawa T, Kurokawa TH et al. Factors affecting the in vitro performance of dentin-bonding systems. Japanese Dental Science Review. 2012;48:53-60. https://doi.org/10.1016/j.jdsr.2011.11.002
13. Perdigão J. Dentin bonding-variables related to the clinical situation and the substrate treatment. Dent Mater. 2010;26(2):e24-37. https://doi.org/10.1016/j.dental.2009.11.149
14. Lapinska B, Klimek L, Sokolowski J, Lukomska-Szymanska M. Dentine surface morphology after chlorhexidine application-SEM study. Polymers (Basel). 2018; 10(8):905. https://doi.org/10.3390/polym10080905
15. Lukomska-Szymanska M, Sokolowski J, Lapinska B. Degradation of a hybrid layer-Review of literature. J Stoma. 2017;70(1):88-94.
16. Pashley DH, Tay FR, Breschi L, Tjäderhane L et al. State of the art etch-and-rinse adhesives. Dent Mater. 2011;27(1):1-16. https://doi.org/10.1016/j.dental.2010.10.016
17. Van Meerbeek B, De Munck J, Yoshida Y, Inoue S et al. Buonocore memorial lecture. Adhesion to enamel and dentin: current status and future challenges. Oper Dent. 2003;28(3):215-235. PMID: 12760693.
18. Van Meerbeek B, Inokoshi S, Braem M, Lambrechts P et al. Morphological aspects of the resin-dentin interdiffusion zone with different dentin adhesive systems. J Dent Res. 1992;71(8):1530-1540. https://doi.org/10.1177/00220345920710081301
19. Ferrari M, Goracci G, García-Godoy F. Bonding mechanism of three "one-bottle" systems to conditioned and unconditioned enamel and dentin. Am J Dent. 1997; 10(5):224-230.
20. Unemori M, Matsuya Y, Akashi A, Goto Y et al. Composite resin restoration and postoperative sensitivity: clinical follow-up in an undergraduate program. J Dent. 2001;29(1):7-13. https://doi.org/10.1016/S0300-5712(00)00037-3
21. Camps J, Déjou J, Rémusat M, About I. Factors influencing pulpal response to cavity restorations. Dent Mater. 2000;16(6):432-440. https://doi.org/10.1016/S0109-5641(00)00041-5
22. De Munck J, Luehrs AK, Poitevin A, Van Ende A et al. Fracture toughness versus micro-tensile bond strength testing of adhesive-dentin interfaces. Dent Mater. 2013;29(6):635-44. https://doi.org/10.1016/j.dental.2013.03.010
23. Chowdhury A, Islam R, Alam A, Matsumoto M et al. Variable smear layer and adhesive application: The pursuit of clinical relevance in bond strength testing. Int J Mol Sci. 2019;20(21):5381. https://doi.org/10.3390/ijms20215381
24. Jacker-Guhr S, Sander J, Luehrs AK. How "Universal" is adhesion? Shear bond strength of multi-mode adhesives to enamel and dentin. J Adhes Dent. 2019;21(1):87-95. https://DOI: 10.3290/j.jad.a41974
25. Stape THS, Wik P, Mutluay MM, Al-Ani AAS et al. Selective dentin etching: A potential method to improve bonding effectiveness of universal adhesives. J Mech Behav Biomed Mater. 2018;86:14-22. https://doi.org/10.1016/j.jmbbm.2018.06.015
26. Tsujimoto A, Barkmeier WW, Takamaizawa T, Watanabe H et al. Comparison between universal adhesives and two-step self etch adhesives in terms of dentin bond fatigue durability in self-etch mode. Eur J Oral Sci. 2017;125(3):215-222. https://doi10.1111/eos.12346
27. Daneshkazemi P, Ghasemi A, Daneshkazemi A, Shafiee F. Evaluation of micro shear bonding strength of two universal dentin bondings to superficial dentin by self-etch and etch-and-rinse strategies. J Clin Exp Dent. 2018;10(9):e837-e843. https://doi.org/10.4317/jced.54740
28. Lezaja Zebic M, Dzeletovic B, Miletic V. Microtensile bond strength of universal adhesives to flat versus class I cavity dentin with pulpal pressure simulation. J Esthet Restor Dent. 2018;30(3):240-248. https://doi.org/10.1111/jerd.12363
29. Papadogiannis D, Dimitriadi M, Zafiropoulou M, Gaintantzopoulou MD et al. Universal adhesives: Setting characteristics and reactivity with dentin. Materials (Basel). 2019;12(10):1720. https://doi.org/10.3390/ma12101720
30. Zhang ZY, Tian FC, Niu LN, Ochala K et al. Defying ageing: An expectation for dentine bonding with universal adhesives? J Dent. 2016;45:43-52. https://doi.org/10.1016/j.jdent.2015.11.008
31. Zecin-Deren A, Sokolowski J, Szczesio-Wlodarczyk A, Piwonski I et al. Multi-layer application of self-etch and universal adhesives and the effect on dentin bond strength. Molecules. 2019;24(2):345. https://doi10.3390/molecules24020345
32. Van Meerbeek B, Peumans M, Poitevin A, Mine A et al. Relationship between bond-strength tests and clinical outcomes. Dent Mater. 2010;26(2):e100–e121. https://

doi10.1016/j.dental.2009.11.148
33. Van Landuyt KL, Snauwaert J, De Munck J, Peumans M et al. Systematic review of the chemical composition of contemporary dental adhesives. Biomaterials. 2007;28(26):3757-3785. https://doi.org/10.1016/j.biomaterials.2007.04.044
34. Yousry MM, ElNaga AA, Hafez RM, El-Badrawy W. Microshear bond strength and interfacial morphology of etch-and-rinse and self-etch adhesive systems to superficial and deep dentin. Quintessence Int. 2011;42(9):e96-e106.
35. Yoshihara K, Hayakawa S, Nagoaka N, Okihara T et al. Etching efficacy of self-etching functional monomers. J Dent Res. 2018;97(9):1010-1016. https://doi.org/10.1016/j.jdent.2015.04.003
36. Rosa WL, Piva E, Silva AF. Bond strength of universal adhesives: A systematic review and meta-analysis. J Dent. 2015;43:765-776. https://doi.org/10.1016/j.jdent.2015.04.003
37. Kumagai RY, Hirata R, Pereira PNR, Reis AF. Moist vs over-dried etched dentin: FE-SEM/TEM and bond strength evaluation of resin-dentin interfaces produced by universal adhesives. J Esthet Restor Dent. 2020;32(3):325-332. https://doi.org/10.1111/jerd.12537
38. Choi AN, Lee JH, Son SA, Jung KH et al. Effect of dentin wetness on the bond strength of universal adhesives. Materials (Basel). 2017;10(11):1224. https://doi.org/10.3390/ma10111224
39. Sugimura R, Tsujimoto A, Hosoya Y, Fischer NG et al. Surface moisture influence on etch-and rinse universal adhesive bonding. Am J Dent. 2019;32(1):33-38.
40. Takamizawa T, Imai A, Hirokane E, Tsujimoto A et al. SEM observation of novel characteristic of the dentin bond interfaces of universal adhesives. Dent Mater. 2019;35(12):1791-1804. https://doi.org/10.1016/j.dental.2019.10.006

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Antibiotic indication in endodontics by Colombian dentists with different levels of training: a survey

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ABSTRACT

Aim**:** *This study investigated how Colombian dentists with different academic levels indicate antibiotics with therapeutic purposes in endodontics.* ***Materials and method*****:** *A cross-sectional survey was conducted among 559 dentists in the form of an online questionnaire.* ***Results*****:** *Three hundred and twenty questionnaires were answered (57.2%). There were significant differences among respondents. For irreversible pulpitis, 140 dentists (43.7%) said they prescribe antibiotics (57.5% of general practitioners, 20.1% of specialists and 38.9% of those with Master's and/or PhD degrees), while for symptomatic apical periodontitis, 183 (57.2%) did so (74.1% of general practitioners, 28.4% of specialists and 50.0% of those with Master's and/or PhD degrees) ($p < 0.05$). Amoxicillin was the most frequently prescribed antibiotic, and its association with clavulanic acid was the most often cited for acute periradicular abscess with systemic involvement.* ***Conclusions*****:** *The greatest misunderstandings in prescribing antibiotics occurred among general practitioners. Considering all clinical conditions that do not require antibiotics, 60% of general practitioners and 34% of specialists, on average, indicated antibiotics.*

Keywords: *antimicrobial stewardship - dental pulp disease - bacteria - dental infection control - antibacterial drug resistance*

Indicação dos antibióticos em Endodontia por dentistas colombianos com diferentes níveis de formação: uma pesquisa por questionário

RESUMO

Objetivo**:** *Este estudo investigou como dentistas colombianos com diferentes níveis acadêmicos indicaram antibióticos com fins terapêuticos em Endodontia.* ***Materiais e método*****:** *Realizou-se um levantamento transversal com 559 dentistas. Foi enviado um questionário online.* ***Resultados*****:** *Foram respondidos 320 questionários (57,2%). Houve diferenças significativas entre os profissionais com diferentes níveis de formação. Para pulpite irreversível, 140 (43,7%) dentistas afirmaram indicar antibióticos (57,5% clínicos gerais, 20,1% especialistas e 38,9% com mestrado e/ou doutorado), enquanto para periodontite apical sintomática, 183 (57,2%) prescrevem estes medicamentos (74,1% clínicos, 28,4% especialistas e 50,0% com mestrado e doutorado) ($p < 0,05$). A amoxicilina foi a mais indicada entre os profissionais, e sua associação com ácido clavulânico foi a mais referida para abscesso perirradicular agudo com acometimento sistêmico.* ***Conclusões*****:** *Os maiores equívocos na prescrição de antibióticos ocorreram com os clínicos gerais. Considerando todas as condições clínicas que não requerem antibióticos, 60% dos clínicos gerais e 34% dos especialistas, em média, indicaram estes medicamentos.*

Palavras-chave: *administração de antimicrobianos - doença da polpa dentária - bactérias - controle de infecção dentária - resistência a medicamentos antibacterianos*

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INTRODUCTION

The discovery and large-scale use of antibiotics since the middle of last century has enormous impact on the treatment of infections, leading to the survival of thousands of people who would otherwise have died, mainly during the second world war[1]. Due to advances in molecular methods in microbiology, especially at the beginning of the current century, "new pathogens" have been detected in different types of infections that affect humans. However, the development, approval, and launch of new antibiotics have not kept pace with this evolution, and several "new microorganisms" already have multiple resistance to traditional antimicrobials[2].

The emergence and spread of antibiotic-resistant pathogens have become important public health problems, requiring global action from the different health areas[3]. It is estimated that infectious diseases will be the main cause of human mortality in the coming decades, mainly due to the growing number of microorganisms that are multi-resistant to antimicrobials[4].

A broad range of bacterial resistance genes has been detected through molecular methods in samples obtained directly from infected root canals[5]. Although the presence of a resistance gene in a sample does not necessarily imply phenotypic resistance, proteomics studies have detected the expression of resistance factors such as TetR and Beta-lactamase in endodontic infections[6,7]. As the unnecessary use of antibiotics can contribute to selecting these resistant microorganisms, antibiotics should be prescribed with great caution.

The American Association of Endodontics (AAE) and the European Society of Endodontology (ESE) frequently revise the guidelines for endodontists regarding proper antibiotic prescription. There is general consensus that in most clinical endodontic situations, it suffices to provide local treatment with removal or reduction of the infection source, without using systemic antibiotics[8-10].

Different studies around the world have shown that dentists still prescribe antibiotics unnecessarily in endodontics[11-14]. There is a clear discrepancy between the recommended protocols for prescribing antimicrobials for patients who really need them and current practices among dentists in different parts of the world[15]. For most Latin American countries, including Colombia, there are few studies on whether antibiotics are prescribed correctly for endodontic purposes. Thus, the aim of this study was to investigate, through an online questionnaire, how professionals with different levels of academic education, who provide endodontic treatment in Colombia, prescribed antibiotics.

MATERIALS AND METHOD

This study was approved by the institutional ethics committee at Universidad Santo Tomas under number 1-18-30082018. A questionnaire (Table 1) was created through Google forms and e-mailed to 559 dentists registered in the Federation of Colombian Dentistry database. The answers were received from February 21 to November 13, 2018. The questionnaire enquired about gender, age, length of professional experience, workplace, region of the country, weekly mean number of patients, monthly mean number of antibiotic prescriptions, clinical situations in which antibiotics are prescribed, duration of antibiotic prescriptions, prescription of loading dose, conduct for patients allergic to penicillins, and management in case of failure of the first-choice antibiotic. The antibiotics listed in the different clinical situations were amoxicillin, amoxicillin with clavulanic acid, azithromycin, cephalexin, clindamycin, erythromycin, penicillin v, and metronidazole.

Sample calculation and statistical analysis

The following parameters were established to estimate the sample size: effect size (w) 0.30 (Cohen's Test scale), power 90%, α error 5%, and degree of freedom equal to 30. Calculation software was G * Power 3.1.9.7 (Universität Kiel, Germany), indicating a total number of 233 individuals. The Kolmogorov-Smirnov and Shapiro-Wilk tests were used to test data normality. The chi-square test or Fisher's exact test were used for the comparison among dentists. The level of statistical significance was established as 5% ($p < 0.05$).

RESULTS

A total 320 (57.2%) questionnaires were answered, 193 (60.3%) by general practitioners, 109 (34.1%) by specialists, and 18 (5,6%) by professionals with Master's or PhD degrees. Most dentists who answered the questionnaire were ≥ 25 years old (n = 269; 84.1%), women (61.6%), and had more than 10 years of professional experience in dentistry (48.4%) (Table 2).

Table 1. Questionnaire about the prescription of antibiotics in endodontics by Colombian dentists

1. What is your level of education in dentistry?

( ) General clinician ( ) Specialist in endodontics ( ) Magister (MSc) ( ) Doctor (DSc / PhD)

2. Gender:

( ) Male ( ) Female

3. Age:

( ) Less than 25 years ( ) 25-35 years ( ) 36-45 years ( ) More than 45 years

4. How long have you been a dental professional?

( ) 0-5 years ( ) 6-10 years ( ) More than 10 years

5. If you are an endodontist, how many years' experience do you have in endodontics only?

( ) 0-5 years ( ) 6-10 years ( ) More than 10 years ( ) I am not an endodontist

6. Where do you conduct your dental practice?

( ) Private practice ( ) Hospital ( ) Academic Institution Clinic ( ) Military Institution ( ) Others

7. In which region of the country do you work?

( ) Southeast ( ) South ( ) Center-west ( ) Northeast ( ) North

8. What is the average number of patients you see per week?

( ) 5-10 ( ) 11-20 ( ) More than 20

9. How many times a month do you prescribe antibiotics, on average?

( ) 1-3 ( ) 4-6 ( ) More than 7 ( ) 0

10. Situations in which you prescribe an antibiotic as an adjunct to treatment for adult patients

	Amoxicillin	Amoxicillin + clavulanic acid	Azithromycin	Cephalexin	Clindamycin	Erythromycin	Penicillin V	Metronidazole	None
Irreversible pulpitis									
Irreversible pulpitis with symptomatic apical periodontitis									
Pulp necrosis									
Symptomatic acute apical periodontitis									
Chronic apical abscess									
Acute apical abscess with localized intraoral edema/ pain									
Acute apical abscess with diffuse intraoral and extraoral edema, fever, and trismus									
Avulsion									
Postoperative pain (after instrumentation/ filling)									
Root perforation									

Table 2. Demographics, academic education levels, and profile of the service provided

Variable	%
Age	
< 25 years	15.9
25-35 years	33.8
36-45 years	22.2
>45 years	28.1
Gender	
Female	61.6
Male	38.4
Academic education level	
General Practitioner	60.3
Specialist in Endodontics	34.1
Master/PhD	5.6
Years' experience	
0-5 years	36.6
6-10 years	15.0
>10 years	48.4
Setting	
Private office	56.9
University	20.3
Hospital	5.3
Military Institution	1.6
Other	15.9
Region of the country	
North	7.2
South	8.1
Northeast	39.1
Midwest	42.5
Southeast	3.1
Mean number of patients per week	
5-10 patients	41.9
11-20 patients	25.6
>20 patients	32.5

Regarding the prescription of antibiotics, 257 professionals (80.3%) prescribe antibiotics for 7 days, and only 6 (1.9%) suspend the prescription after the symptoms disappear. Comparison among the groups for prescription time showed no statistical difference ($p > 0.05$), suggesting that the level of training does not influence this decision (Table 3). More than half of the respondents prescribe antibiotics in up to 3 cases per month, and only 7% do not prescribe them. Regardless of the clinical situation and the professional training, amoxicillin is the most frequently prescribed antibiotic. Only in acute periradicular abscess with systemic involvement, the association of amoxicillin with clavulanic acid was the most frequently reported prescription. Also, 243 professionals (75.9%) responded that they do not prescribe an attack dose. Clindamycin was the most frequently recommended antibiotic in case of allergy to penicillin (n = 120; 37.5%), followed by erythromycin (n = 97; 30.3%) and azithromycin (n = 76; 23.8%).

Comparing the three groups of professionals, only the variable "2nd choice in case of allergy to penicillin" showed a significant difference ($p < 0.01$), with specialists in endodontics presenting the highest frequency of clindamycin prescription (54.1%). In the other groups, Master's or PhD and clinicians, azithromycin (44.4%) and erythromycin (37.8%) were the most frequently prescribed alternatives. When the antibiotic does not have the desired effect, 52.5% of the professionals choose to change the antibiotic, without significant differences among groups.

There were significant differences in antibiotic prescription among groups for irreversible pulpitis with symptomatic apical periodontitis and symptomatic acute apical periodontitis ($p < 0.01$). For irreversible pulpitis with symptomatic apical periodontitis, 140 dentists (43.7%) said they

Table 3. Comparison among the three levels of academic education for general clinical conduct in antibiotic prescription

Variable	Academic Education Level				*p*-value
	General Practitioner (N = 193) N (%)	Specialist in Endodontics (N = 109) N (%)	Master/PhD (N = 18) N (%)	Total (N = 320) N (%)	
Prescription time					0.422
3 days	9 (4.7)	6 (5.5)	0 (0.0)	15 (4.7)	
5 days	19 (9.8)	12 (11.0)	5 (27.8)	36 (11.2)	
7 days	158 (81.9)	87 (79.8)	12 (66.7)	257 (80.3)	
10 days	4 (2.1)	1 (0.9)	0 (0.0)	5 (1.6)	
14 days	1 (0.5)	0 (0.0)	0 (0.0)	1 (0.3)	
Until symptoms disappear	2 (1.0)	3 (2.8)	1 (5.5)	6 (1.9)	

prescribe antibiotics (57.5% general practitioners, 20.1% specialists, and 38.9% with Master's or PhD degrees), while for symptomatic acute apical periodontitis, 183 (57.2%) prescribe antibiotics (74.1% clinical, 28.4% specialists and 50.0% with Master and PhD). In cases of abscesses, clinicians prescribe significantly more antibiotics than specialists and professionals with Master's or PhD degrees, not only for chronic cases (78.2%, 45.9%, and 50%, respectively, $p < 0.001$), but also for acute cases with localized intraoral edema/pain (94.3%, 87.2%, and 66.7%, respectively, $p < 0.05$). In cases of root perforation, 61.1% of clinicians prescribe some antibiotic, against 26.6% of specialists and 50% of professionals with Master's or PhD degrees ($p < 0.01$).

DISCUSSION

Healthcare professionals often use systemic antimicrobials to treat or prevent infections. However, there is still a global threat to the effectiveness of these agents related to their indiscriminate use, resulting in the emergence of resistant microorganisms[16]. This concern also applies to endodontics, since antibiotic resistance by bacteria isolated from infected root canals has been frequently reported[17]. Moreover, the number of deaths related to endodontic infections refractory to antibiotic treatment is significant[18].

Endodontic infections are polymicrobial, which means that multiple species and virulence factors are involved[7]. Sometimes, the immune system cannot suppress this type of infection, and antibiotics are required[18]. However, the prescription of antibiotics in endodontics should be limited to certain clinical conditions, with the aim of preventing the spread of infection and the development of secondary infections in medically compromised patients. As verified in the present study, many professionals lack knowledge about the proper use of antibiotics in endodontics. In this context, the main contribution of this study was to recognize and point out the magnitude of this problem among Colombian dentists.

The present study was based on a questionnaire about antibiotic prescription in different clinical situations, which was answered by 320 dentists with different levels of training. Studies using similar questionnaires have been conducted in different countries[11,12,14,19,20]. However, this was the first study to investigate antibiotic prescription habits among dentists in Colombia. The response rate was acceptable (54.7%), compared to similar studies in Spain (31.1%)[11], Norway (27.2%)[19], United States (22.9%)[20], and Brazil (4.4%)[14].

In this type of study, it is important to record the level of professional training and geographic location in order to design continuing education strategies, if necessary. The current study compared different regions of Colombia, but found no significant difference among them regarding prescription or level of professional training.

The greatest misunderstandings in prescribing antibiotics occurred among general practitioners. Considering all clinical conditions that do not require antibiotics, 60% of general practitioners, on average, prescribed them, while only 34% of endodontics specialists did so. The mean for professionals with Master's or PhD degrees was 45%, which contradicts expectations, considering their higher level of education. Another alarming finding was for avulsion conditions, which do require antibiotic therapy, but for which 44% of general practitioners reported they did not prescribe antibiotics. The same occurred with 25% of specialists and 39% of professionals with Master's and PhD degrees. Specific training in endodontics is the most likely explanation for the fact that specialists prescribe more accurately and better than other professionals.

In the present study, professionals with different levels of education prescribe antibiotics for irreversible pulpitis (21.2%) and irreversible pulpitis with symptomatic apical periodontitis (43.7%), which is a matter of concern. Dentists prescribe antibiotics to reduce the patient's pain, though there is no evidence in the literature justifying it[21]. In irreversible pulpitis with acute apical periodontitis, the pulp remains vital, with no infection or signs and symptoms of systemic involvement. In these cases, there is only an inflammatory process in the pulp, and therefore, antibiotics are not indicated[21]. The level of professional training showed a statistical difference in this case (Table 4). A low percentage was found for this situation in studies in other countries such as Lithuania (19.4%)[12] and Brazil (6.2%)[14], while the percentage was higher in a study in India[22]. The significant difference between the results of the present study and those observed in Lithuania[12] and Brazil[14] may be related to the fact that the present study included general practitioners

Table 4. Frequency of antibiotic prescription in clinical conditions according to different academic education levels

Variable	Academic Education Level				*p*-value
	General Practitioner	Specialist in Endodontics	Master/PhD	Total	
	(N = 193)	(N = 109)	(N = 18)	(N = 320)	
	N (%)	N (%)	N (%)	N (%)	
Irreversible pulpitis					0.052
Prescribed antibiotics	54 (28)	10 (9.1)	4 (22.2)	68 (21.2)	
No	139 (72)	99 (90.9)	14 (77.8)	252 (78.8)	
Irreversible pulpitis with symptomatic apical periodontitis					< 0.001
Prescribed antibiotics	111 (57.5)	22 (20.1)	7 (38.9)	140 (43.7)	
No	82 (42.5)	87 (79.9)	11 (61.1)	180 (56.3)	
Pulp necrosis					0.19
Prescribed antibiotics	86 (44.6)	25 (22.9)	6 (33.4)	117 (36.5)	
No	107 (55.4)	84 (77.1)	12 (66.6)	203 (63.5)	
Symptomatic acute apical periodontitis					< 0.0001
Prescribed antibiotics	143 (74.1)	31 (28.4)	9 (50.0)	183 (57.2)	
No	50 (25.9)	78 (71.6)	9 (50.0)	137 (42.8)	
Chronic apical abscess					0.0001
Prescribed antibiotics	151 (78.2)	50 (45.9)	9 (50.0)	210 (65.6)	
No	42 (21.8)	59 (54.1)	9 (50.0)	110 (34.4)	
Acute apical abscess with localized intraoral edema/pain					0.045
Prescribed antibiotics	182 (94.3)	95 (87.2)	12 (66.7)	289 (90.4)	
No	11 (5.7)	14 (12.8)	6 (33.3)	31 (9.6)	
Acute apical abscess with diffuse intra and extraoral edema, fever, and trismus					0.025
Prescribed antibiotics	186 (96.4)	107 (98.2)	15 (83.4)	308 (96.2)	
No	7 (3.6)	2 (1.8)	3 (16.6)	12 (3.8)	
Avulsion					0.077
Prescribed antibiotics	108 (56)	82 (75.2)	11 (61.1)	201 (62.9)	
No	85 (44.0)	27 (24.8)	7 (38.9)	119 (37.1)	
Postoperative pain (after instrumentation/filling)					0.408
Prescribed antibiotics	78 (43.1)	31 (28.4)	8 (45.5)	117 (36.6)	
No	115 (59.6)	78 (71.6)	10 (55.5)	203 (63.4)	
Root perforation					< 0.0001
Prescribed antibiotics	118 (61.1)	29 (26.6)	9 (50.0)	156 (48.7)	
No	75 (38.9)	80 (73.4)	9 (50.0)	164 (51.3)	

who perform endodontic treatment, while the other studies included only specialists in endodontics.

Due to the short half-life of antibiotics, a minimum serum inhibitory concentration is essential for the success of antibiotic therapy. Therefore, a higher initial dose (attack dose) is usually recommended to ensure antibiotic penetration into bone tissue in a concentration high enough to eliminate the microorganisms in the infection site. The present findings showed that more than 70% of

all interviewed professionals do not prescribe attack doses. Different results were found in other countries[14,20], where most professionals do prescribe the attack dose.

Regarding the second-choice antibiotic in cases of penicillin allergy, most professionals choose clindamycin (37.5%), regardless of training level. A similar rate was found in Brazil (33%)[14]. However, differences were found in intragroup analysis in which professionals with Master's or PhD degrees prescribe more azithromycin; specialists prescribe more clindamycin; and clinicians prescribe more erythromycin. Current recommendations suggest azithromycin instead of clindamycin in these cases[23]. The problem with clindamycin is the risk of infection by *Clostridioides difficile*, and the consequent development of pseudomembranous colitis. It is important to emphasize that not only clindamycin, but also many other broad-spectrum antibiotics have been associated with this type of adverse reaction, and the risk increases with longer treatment periods and greater number of antibiotics administered[24].

Systemic antibiotics are unnecessary in most cases in endodontics, including acute abscesses located without systemic involvement[18,25]. However, antibiotic therapy is a crucial coadjuvant to treat cases of cellulite with signs of systemic effects, such as lymphadenopathy, limited mouth opening (trismus), fever, loss of appetite, and general malaise. These symptoms suggest that the patient's immune system is not controlling the infection, and the microorganisms may spread to other anatomical spaces[23]. In the present study, most respondents prescribe antibiotics for acute periradicular abscesses with systemic involvement, although the prescription frequency was lowest among the professionals with higher qualifications (Table 4). In these most critical situations, respondents' antibiotic of choice was amoxicillin associated with clavulanic acid, as has been recommended[23]. The association of these two drugs provides a greater spectrum of action, including penicillin-resistant strains.

It is clear that clinicians should keep in mind that antibiotics should be indicated as a therapeutic adjunct to local treatment to help prevent the spread of infection in severe cases[9]. Continuing medical and dental education should be encouraged in order to improve the indication of systemic antimicrobials and prevent erroneous prescriptions, reducing the probability of development of bacterial resistance[15]. There is a clear need to improve antibiotic recommendations as well as knowledge of pulpal and periradicular diseases, which are a problem not only locally, but also globally.

A limitation of the present study is that the professionals were not asked about why they did or did not prescribe antibiotics, as was done in a previous study[20]. Such answers could help identify the degree of information or misinformation about the prescription of antibiotics. However, the longer the questionnaire, the less accurate the information provided[26].

CONCLUSIONS

The greatest misunderstandings in prescribing antibiotics occurred among general practitioners. Considering all clinical conditions that do not require antibiotics, 60% of general practitioners, on average, indicated antibiotics, while only 34% of specialists in endodontics did so. This information reinforces the need to create continuing education programs for Colombian dentists, in order to avoid unnecessary prescriptions, thereby reducing the development of microbial resistance to antibiotics.

DECLARATION OF CONFLICTING INTERESTS

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Mohr KI. History of Antibiotics Research. Curr Top Microbiol Immunol. 2016;398:237-272. https://doi.org/10.1007/82_2016_499
2. Bumann D. Pathogen proteomes during infection: A basis for infection research and novel control strategies. J Proteomics. 2010;73(11):2267-2276.https://doi.org/10.1016/j.jprot.2010.08.004.
3. Gajdács M. The Concept of an Ideal Antibiotic: Implications for Drug Design. Molecules. 2019 Mar 3;24(5):892.https://doi.org/10.3390/molecules24050892.
4. Weiner LM, Webb AK, Limbago B, Dudeck MA*, et al.* Antimicrobial-Resistant Pathogens Associated With Healthcare-Associated Infections: Summary of Data Reported to the National Healthcare Safety Network at the Centers for Disease Control and Prevention, 2011-2014. Infect Control Hosp Epidemiol. 2016;37(11):1288-1301. https://doi.org/10.1017/ice.2016.174.
5. Rocas IN, Siqueira JF, Jr. Antibiotic resistance genes in anaerobic bacteria isolated from primary dental root canal infections. Anaerobe. 2012;18(6):576-580.https://doi.org/10.1016/j.anaerobe.2012.10.001.
6. Nandakumar R, Madayiputhiya N, Fouad AF. Proteomic analysis of endodontic infections by liquid chromatography-tandem mass spectrometry. Oral Microbiol Immunol 2009;24(4):347-352.https://doi.org/10.1111/j.1399-302X.2009.00520.x.
7. Provenzano JC, Siqueira JF, Jr., Rocas IN, Domingues RR*, et al.* Metaproteome analysis of endodontic infections in association with different clinical conditions. PLoS One. 2013;8(10):e76108.https://doi.org/10.1371/journal.pone.0076108.
8. AAE position statement: AAE guidance on the use of systemic antibiotics in endodontics. J Endod. 2017;43(9):1409-1413. https://doi.org/10.1016/j.joen.2017.08.015.
9. Segura-Egea JJ, Gould K, Sen BH, Jonasson P*, et al.* European Society of Endodontology position statement: the use of antibiotics in endodontics. Int Endod J. 2018;51(1):20-25.https://doi.org/10.1111/iej.12781.
10. Abraham SB, Abdulla N, Himratul-Aznita WH, Awad M*, et al.* Antibiotic prescribing practices of dentists for endodontic infections; a cross-sectional study. PLoS One. 2020;15(12):e0244585.https://doi.org/10.1371/journal.pone.0244585.
11. Rodriguez-Nunez A, Cisneros-Cabello R, Velasco-Ortega E, Llamas-Carreras JM*, et al.* Antibiotic use by members of the Spanish Endodontic Society. J Endod. 2009;35(9):1198-1203.https://doi.org/10.1016/j.joen.2009.05.031.
12. Skucaite N, Peciuliene V, Maneliene R, Maciulskiene V. Antibiotic prescription for the treatment of endodontic pathology: a survey among Lithuanian dentists. Medicina (Kaunas). 2010;46(12):806-813
13. Alattas HA, Alyami SH. Prescription of antibiotics for pulpal and periapical pathology among dentists in southern Saudi Arabia. J Glob Antimicrob Resist. 2017;9:82-84. https://doi.org/10.1016/j.jgar.2017.01.012.
14. Bolfoni MR, Pappen FG, Pereira-Cenci T, Jacinto RC. Antibiotic prescription for endodontic infections: a survey of Brazilian Endodontists. Int Endod J. 2018;51(2):148-156.https://doi.org/10.1111/iej.12823.
15. Shemesh A, Batashvili G, Shuster A, Slutzky H*, et al.* International questionnaire study on systemic antibiotics in endodontics. Part 1. Prescribing practices for endodontic diagnoses and clinical scenarios. Clin Oral Investig. 2022;26(3):2921-2926.https://doi.org/10.1007/s00784-021-04274-z.
16. Kandasamy G, Sivanandy P, Almaghaslah D, Khobrani M*, et al.* Knowledge, attitude, perception and practice of antibiotics usage among the pharmacy students. Int J Clin Pract 2020:e13599.https://doi.org/10.1111/ijcp.13599.
17. Jungermann GB, Burns K, Nandakumar R, Tolba M*, et al.* Antibiotic resistance in primary and persistent endodontic infections. J Endod. 2011;37(10):1337-1344.https://doi.org/10.1016/j.joen.2011.06.028.
18. Siqueira JF, Jr., Rocas IN. Microbiology and treatment of acute apical abscesses. Clin Microbiol Rev. 2013;26(2):255-273.https://doi.org/10.1128/CMR.00082-12.
19. Preus HR, Fredriksen KW, Vogsland AE, Sandvik L*, et al.* Antibiotic-prescribing habits among Norwegian dentists: a survey over 25 years (1990-2015). Eur J Oral Sci. 2017;125(4):280-287.https://doi.org/10.1111/eos.12360.
20. Germack M, Sedgley CM, Sabbah W, Whitten B. Antibiotic Use in 2016 by Members of the American Association of Endodontists: Report of a National Survey. J Endod. 2017;43(10):1615-1622.https://doi.org/10.1016/j.joen.2017.05.009.
21. Agnihotry A, Thompson W, Fedorowicz Z, van Zuuren EJ*, et al.* Antibiotic use for irreversible pulpitis. Cochrane Database Syst Rev. 2019;5:CD004969.https://doi.org/10.1002/14651858.CD004969.pub5.
22. Garg AK, Agrawal N, Tewari RK, Kumar A*, et al.* Antibiotic prescription pattern among Indian oral healthcare providers: a cross-sectional survey. J Antimicrob Chemother. 2014;69(2):526-528.https://doi.org/10.1093/jac/dkt351.
23. Lockhart PB, Tampi MP, Abt E, Aminoshariae A*, et al.* Evidence-based clinical practice guideline on antibiotic use for the urgent management of pulpal- and periapical-related dental pain and intraoral swelling: A report from the American Dental Association. J Am Dent Assoc. 2019;150(11):906-921 e912.https://doi.org/10.1016/j.adaj.2019.08.020.
24. Guh AY, Kutty PK. Clostridioides difficile Infection. Ann Intern Med. 2018;169(7):ITC49-ITC64.https://doi.org/10.7326/AITC201810020.
25. Fouad AF, Rivera EM, Walton RE. Penicillin as a supplement in resolving the localized acute apical abscess. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 1996;81(5):590-595.https://doi.org/10.1016/s1079-2104(96)80054-0.
26. Kost RG, de Rosa JC. Impact of survey length and compensation on validity, reliability, and sample characteristics for Ultrashort-, Short-, and Long-Research Participant Perception Surveys. J Clin Transl Sci. 2018;2(1):31-37.https://doi.org/10.1017/cts.2018.18.

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Bonding strategy of a universal adhesive system containing chitosan: influence on dentin permeability, and effect on adhesive layer micromorphology

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ABSTRACT

Aim: *This study evaluated the influence of chitosan added to a universal adhesive system used in total-etch (TE) or self-etch (SE) mode on dentin permeability, and on the micromorphology of the adhesive layer.* ***Materials and Method:*** *Dentin discs were obtained from human third molars and randomly distributed according to bonding strategy (TE or SE), and to whether or not 1% chitosan (C) was added to a universal adhesive system (Single Bond Universal/3M ESPE), to create the following groups (n=10): TE, TEC, SE, and SEC. Dentin permeability was measured at baseline and after application of dentin treatments. The surface of the adhesive layer (AL) and the dentin adjacent to the AL were examined under a scanning electron microscope.* ***Results:*** *There were no significant differences in permeability percentage between the groups with and without C (TE and SE versus TEC and SEC) (p>0.05; Mann Whitney test). Dentin permeability was lower when the adhesive system was applied in the SE mode, regardless of the addition of C. The micromorphology of the AL surface showed irregularities, and a greater degree of porosity, when the adhesive system was applied in the SE mode, regardless of chitosan addition. There was a greater depth of penetration of the adhesives into the dentin adjacent to the AL in both the TE and TEC groups. Chitosan added to the adhesive system did not influence dentin permeability.* ***Conclusions:*** *The self-etch strategy led to lower dentin permeability, and to more irregularities on the surface of the adhesive layer.*

Keywords: *adhesive system - chitosan - dentin permeability - scanning electron microscopy*

Estratégia de união de um sistema adesivo universal contendo quitosana: influência na permeabilidade dentinária e efeito na micromorfologia da camada do adesivo

RESUMO

Objetivo: *Este estudo avaliou a influência da quitosana adicionada a um sistema adesivo universal usado no modo total-etch (TE) ou self-etch (SE) na permeabilidade dentinária e na micromorfologia da camada adesiva.* ***Materiais e método:*** *Discos de dentina foram obtidos de terceiros molares humanos e distribuídos aleatoriamente de acordo com a estratégia de união (TE ou SE), e para incorporação ou não de quitosana a 1% (Q) em um sistema adesivo universal (Single Bond Universal/3M ESPE), para obter os seguintes grupos (n=10): TE, TEQ, SE e SEQ. A permeabilidade da dentina foi medida no início e após a aplicação dos tratamentos de dentina. A superfície da camada adesiva (CA) e a dentina adjacente à CA foram examinadas em microscópio eletrônico de varredura.* ***Resultados:*** *Não houve diferenças significativas no percentual de permeabilidade entre os grupos com e sem Q (TE e SE versus TEQ e SEQ) (p>0,05; teste de Mann Whitney). Houve um menor percentual de permeabilidade dentinária quando o sistema adesivo foi aplicado no modo SE, independentemente da incorporação de Q. A micromorfologia da superfície da CA apresentou irregularidades e maior grau de porosidade quando o sistema adesivo foi aplicado no modo SE, independentemente da adição de Q. Houve maior profundidade de penetração dos adesivos na dentina adjacente à CA nos grupos TE e TEQ. A quitosana adicionada ao sistema adesivo não influenciou a permeabilidade dentinária.* ***Conclusões:*** *A estratégia autocondicionante levou a uma menor permeabilidade dentinária e a mais irregularidades na superfície da camada adesiva.*

Palavras-chave: *sistema adesivo - quitosana - permeabilidade dentinária - microscopia eletrônica de varredura*

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INTRODUCTION

Dentin hypersensitivity is characterized by pain caused by exposure of dentin due to opening of the dentinal tubules, and displacement of intratubular fluid. The pain caused by this inner fluid movement[1] can be exacerbated by thermal, tactile, or chemical stimuli[2]. Different treatment strategies have been proposed to minimize hypersensitivity. One approach is to apply tubular occlusion agents to the dentin surface as a mechanical barrier to hinder the movement of dentinal fluid[1].

Adhesive systems promote the formation of a hybrid layer by micromechanical retention between dentinal collagen fibrils[3]. Fu et al.[4] observed that adhesives effectively occluded dentinal tubules, thereby significantly reducing dentin permeability. Dentin permeability is reduced regardless of whether total-etching, self-etching or universal adhesive systems are used[5]. However, an adhesive system using a total-etching strategy -requiring prior acid etching- can open dentinal tubules, increasing their diameter, and immediately exacerbating pre-existing pain[6]. In addition, there is a discrepancy between the depth of demineralization from acid use and the penetration capacity of the adhesive due to its molecular size[7]. Self-etching adhesive systems eliminate the step of total removal of the smear layer, and reduce the clinical steps required.

Adding components to the adhesive systems enables deposition of products on the dentin surface or at the entrance of the dentinal tubules, thereby reducing dentin permeability and sensitivity. Chitosan has been evaluated for its interactive effect with dentin collagen to stabilize the dentin matrix[8,9]. It is deposited on the surface and inside the dentinal tubules when applied as a rewetting agent, and it forms a calcium phosphate layer on the demineralized dentin layer[10]. This effect may reduce dentin permeability and hypersensitivity, regardless of the removal of the smear layer.

The addition of chitosan to total-etching and self-etching adhesive systems has been evaluated for its antimicrobial effect in concentrations of 0.12 to 5%[10,11], and to determine any interference regarding bond strength to dentin[8], or possible improvement in the quality of adhesion[10]. However, none of the concentrations tested has been found to improve these properties. Chitosan added to an adhesive system may promote remineralization[10], especially remineralization of deep cavities, thereby reducing permeability and pulp cytotoxicity[12]. It is therefore important to analyze the influence of chitosan on dentin permeability if it is added to a universal adhesive system used in total-etching or self-etching mode. Thus, the null hypotheses were that there is no difference regarding dentin permeability: H01) when chitosan is added to a universal adhesive system, regardless of whether total-etching or self-etching bonding strategy is used; and H02) when a universal adhesive system is used in total-etching or self-etching strategy.

MATERIALS AND METHOD

Adhesive system specifications and preparation

The materials used in the study are specified in Table 1. The adhesive system with added chitosan was prepared by weighing chitosan PA (low molecular weight Chitosan 448869) on a precision analytical balance (XPR10, Mettler-Toledo GmbH, Greifensee, Switzerland), and adding it to the single bond universal (3M ESPE, St. Paul, MN, USA) adhesive system at a concentration of 1%. Each solution was vortexed vigorously (Phoenix, AP-56, Araraquara, SP, Brazil) for 180 seconds, and stored in a hermetically closed flask, isolated from light and moisture to eliminate bubbles. The pH of the adhesive system, whether or not chitosan was added, was measured in triplicate with a microelectrode coupled to a pH meter (pH meter TEC-2/ Tecnal/ Piracicaba, SP, Brazil) (Table 1).

Tooth selection and dentin disc preparation

The project was approved by the Research Ethics Committee of the Faculdade São Leopoldo Mandic (CAAE 26445019.7.0000.5374). Forty sound third molars, which had been extracted, stored, and frozen (for a maximum period of six months) were selected. After the teeth were cleaned with periodontal curettes and scalpel blades, they were sectioned using a precision metallographic cutter (Isomet 1000, Buehler, Springfield, VA, USA), with a high-concentration diamond disc (Buehler, 102mm x 0.3mm) under cooling, to obtain dentin discs. The crowns were sectioned perpendicular to the long axis of the tooth, 1.5 mm above the cementoenamel junction[13], to obtain discs approximately 1.7-mm. thick (+0.1 mm), and samples with a completely enamel-free occlusal surface.

A small diamond bur marking was made to identi-

Table 1. Specifications of materials used in the study.

Materials (Manufacturer, city, state, country)/Lot	Composition (% by weight) / pH	Mode of use
Single Bond Universal (3M ESPE, Saint Paul, MN, USA) / Lot 1932900226	2-Hydroxyethyl methacrylate (15 - 25), bisphenol A diglycidyl ether dimethacrylate (BisGMA) (15 - 25), ethanol (10 - 15), water 10 - 15), 1,10-decanediol phosphate methacrylate (1 - 10) , copolymer of acrylic and itaconic acids (1 - 5), camphorquinone (< 2) and N,N-dimethylbenzocaine (< 2) */ pH = 2.71	Apply the adhesive for 20 seconds (active application); apply a soft jet of air over the adhesive for 5 seconds; photoactivate for 10 seconds.
Ultra-Etch 35% (Ultradent Products, South Jordan, Utah, USA) Lot BHP7V	Phosphoric Acid (<40%), Aluminum cobalt (<1%); pH = 1.02	Apply phosphoric acid for 15 seconds, wash for 20 seconds, remove excess water and leave moist with absorbent paper
Chitosan 448869 (Sigma Aldrich, St. Louis, MO, USA)	Low molecular weight chitosan (deacetylated chitin) / pH = 2.70	Added to the universal adhesive system at 1%

* According to the MSDS provided by the manufacturer

fy the occlusal surface. Next, both sides of the disc were sanded with 600-grit sandpaper (Lixa de Água, 3M, 3M do Brasil, Sumaré, SP, Brazil) to obtain specimens with uniform, smooth surfaces 1.5-mm thick. The thickness of each specimen was checked with a digital caliper (Mitutoyo Sul Americana LTDA, MIP/E – 103, Suzano, SP, Brazil). The discs were washed with water, and stored in J10 flasks with 5 mL of distilled water for 24 hours.

Both surfaces of the discs were submitted to acid-etching (Ultra-Etch 35%, Ultradent Products, South Jordan, UT, USA) for 15 seconds, to promote opening of the dentinal tubules, and measure the initial dentin permeability values. This served as the first measurement of hydraulic conductance, considered 100% maximum filtration. Then, the occlusal surfaces of the specimens were sanded with 600 grit sandpaper for 30 seconds to form a standardized smear layer.

The 40 dentin discs were randomly divided into four groups (n=10), according to the type of adhesive strategy (Total-Etch/TE or Self-Etch/SE), and addition (C) or non-addition of chitosan to the adhesive system:

- TE: application of 35% phosphoric acid for 15 seconds, and application of a universal adhesive system
- TEC: application of 35% phosphoric acid for 15 seconds, and application of a universal adhesive system containing 1% chitosan
- SE: application of a universal adhesive system
- SEC: application of a universal adhesive system containing 1% chitosan

The adhesive system (with or without chitosan) was applied according to the manufacturer's instructions (Table 1). When the total-etch strategy was used, phosphoric acid was applied for 15 seconds, followed by washing for 20 seconds, and drying with absorbent paper strips. Photoactivation was performed for 10 seconds using a photoactivating device (Valo Cordless, LED, Ultradent Products, South Jordan, UT, USA) with a power of 1,000mW/cm². Next, the second (final) measurement of hydraulic conductance was performed.

Permeability Test

Permeability was evaluated with a device for measuring dentin permeability (THD, Odeme Dental Research, Luzerna, SC, Brazil) under pressure of 5 psi, equivalent to 351.54 cmH_2O. The specimen was mounted in the filtration chamber, and the device adjusted so that water penetrated the dentinal tubules under pressure and was pushed upward toward the occlusal surface. This fluid movement was recorded according to the difference in the position of an air bubble in the glass capillary of the device. Three measurements were taken, the first after 4 minutes, and the next two at 3-minute intervals[14]. The measurements were performed with the digital caliper attached to the permeability measuring device. After the three measurements were taken, the system was depressurized, and the sample dismounted from the chamber. Each dentin disc was repositioned for the final measurement (after the treatments), and mounted in the filtration chamber using a standardized procedure, after

which the system was closed and prepared to take a new measurement. The linear displacement of the liquid in the glass capillary for a preset time was used to calculate the amount of fluid that passed through the specimen by the following formula: $Q = (ri^2l)/t$, where Q ($\mu L/min^{-1}$) is the amount of fluid that passed through the specimen, l (cm) is the linear displacement in the glass capillary, t (min) is the time, and ri (cm^2) is the inner radius of the glass capillary tube.

Hydraulic conductance (L) was calculated using another formula, considering water viscosity and constant specimen thickness: L = Q/(AP), where L is hydraulic conductance ($\mu L\ cm^{-2}\ min\text{-}1\ cmH2O^{-1}$), A ($cm^2$) is the dentin surface area and P (cmH_2O) is the pressure applied. The hydraulic conductance of each dentin disc was evaluated at two time points: initially (after the acid etching) and after the treatments. The percentage of dentin permeability was calculated using the following equation, and each tooth was its own control: L (%) = [(L1-L2) x100]/L1[15], considering L as the permeability percentage, L1 as the hydraulic conductance after removal of the smear layer, and L2 as the hydraulic conductance after application of the treatments.

Analysis by Scanning Electron Microscopy

To determine what material could penetrate deeply into the dentinal tubules, three treated discs were cut sagittally to observe penetration into the dentinal tubules of the adhesive options with or without chitosan. Another three discs from each group were not cut, to be used to evaluate the dentin surface for obliteration or non-obliteration of the dentinal tubules.

The interface of the cut disc sections was polished with sandpaper of decreasing abrasive grit (400, 600, 1200) (Imperial Wetordry, 3M, Sumaré, SP, Brazil). After abundant washing, the samples were demineralized for 30 seconds with hydrochloric acid (HCl), washed again, and subjected to a deproteinizing treatment with 1% sodium hypochlorite solution (Milton's Solution) for 2 min, washed with distilled water for 15 seconds, and dried with absorbent paper[16].

All specimens (sections and uncut discs) were coated in gold for 60 seconds, and examined by Scanning Electron Microscopy (SEM) (Jeol 5900LV, Jeol Ltd, Tokyo, Japan), at a voltage of 15 Kv, with 1000x and 3000x magnification, respectively. The differences in the adhesive penetration observed in the images of the sections and the surface were evaluated qualitatively according to the groups, regarding obliteration or non-obliteration of the dentin surface from the treatments. The images were evaluated by a single examiner previously trained to perform micromorphological evaluations.

Statistical analysis

All the analyses were performed using the R program (R Core Team, 2021). Initially, descriptive and exploratory analyses of the permeability percentage data were performed. The exploratory analysis indicated that the data did not meet the assumptions of parametric analysis, and the Mann Whitney test was applied to compare the two bonding strategies to each other, and with and without chitosan. The significance level was set at 5%.

RESULTS

There was no significant difference in the permeability percentage between the groups with and without chitosan, either for the total-etching (p=0.8206) or the self-etching (p=0.5454) strategies (Table 2). However, there was a significant difference in this percentage between the adhesive system strategies when chitosan was added. Dentin permeability was higher when the total-etching strategy was used, and lower when the self-etching strategy was used, based on the addition (p=0.0002) or non-addition (p=0.0126) of chitosan in the adhesive system (Table 2).

Table 2. Median (minimum and maximum value) of the dentin permeability percentage according to the type of strategy and presence of chitosan in the adhesive system.

Chitosan	Type of strategy	
	TE	SE
Absent	-67.23 (-155.14; -15.98) Ba	26.30 (-125.63; 72.86) Aa
Present	-69.61 (-132.14; 1.66) Ba	34.59 (0.27; 69.07) Aa

Medians followed by different uppercase letters horizontally, and lowercase letters vertically indicate a significant difference (p<0.05).

Micromorphological images of the dentin sections showed that adhesive penetration was deeper for the total-etching bonding strategy (≅ 10 µm) than for the self-etching strategy (≅1 to 3 µm) (Fig. 1), regardless of the presence of chitosan. Application of the adhesive system provided a more intact surface [with fewer irregularities] in the total-etching strategy than in the self-etching strategy, whether with or without chitosan. In the latter application, the surfaces presented more holes and cracks (Fig. 2).

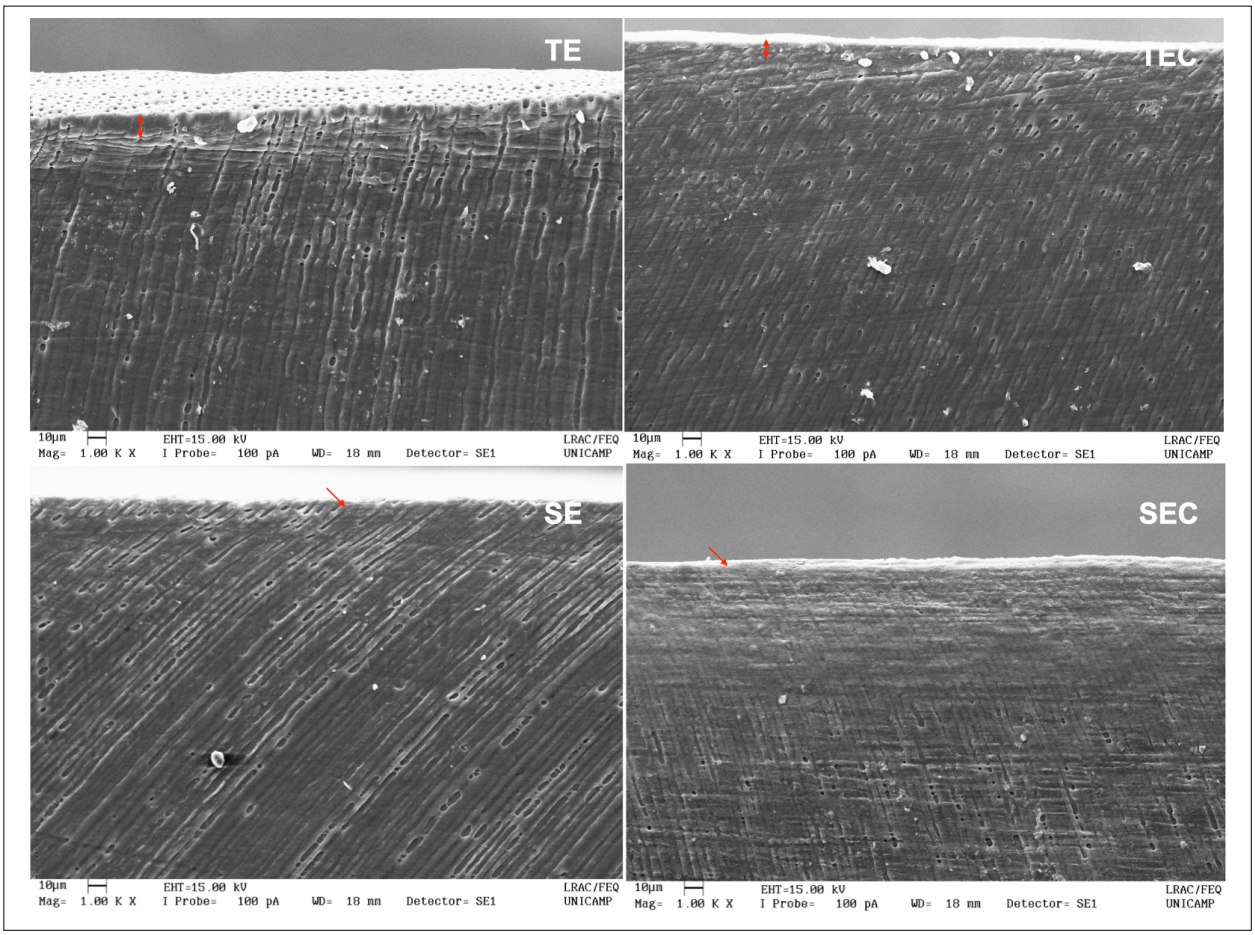

Fig. 1: *Images of the micromorphology of dentin sections (1000x magnification). The red arrows show the penetration depth of the adhesive system. Note that penetration in the total-etch strategy (TE and TEC) was greater, and penetration in the self-etching strategy (SE and SEC) was lower. TE - Total-etch; TEC - Total-etch with chitosan; SE - Self-etching; SEC - Self-etching with chitosan.*

DISCUSSION

There are various components that can be added to dental adhesives, and which could result in lower permeability because they contain particles which, when deposited at the base of dentinal tubules, can block fluid movement. This mechanism of action —explained by Brannstrom's Theory of Hydrodynamics— promotes a reduction in dentin permeability[17]. Hindering the fluid movement in the dentin tubules helps reduce dentin sensitivity. Nevertheless, the addition of chitosan to the universal adhesive system did not influence dentin permeability, regardless of the adhesive strategy, thereby leading to the acceptance of the first null hypothesis of the present study.

Chitosan has an average molecular size of 0.5 µm[7], which enables it to penetrate the dentinal tubules passively, even through the adhesives, considering that the adhesives have a diameter 4 times greater than the average size of chitosan. However, when chitosan is added to the adhesive system, some changes in its physicochemical properties occur[18]. Dacoreggio et al.[18] used a 0.5% concentration of chitosan in the adhesive without causing any

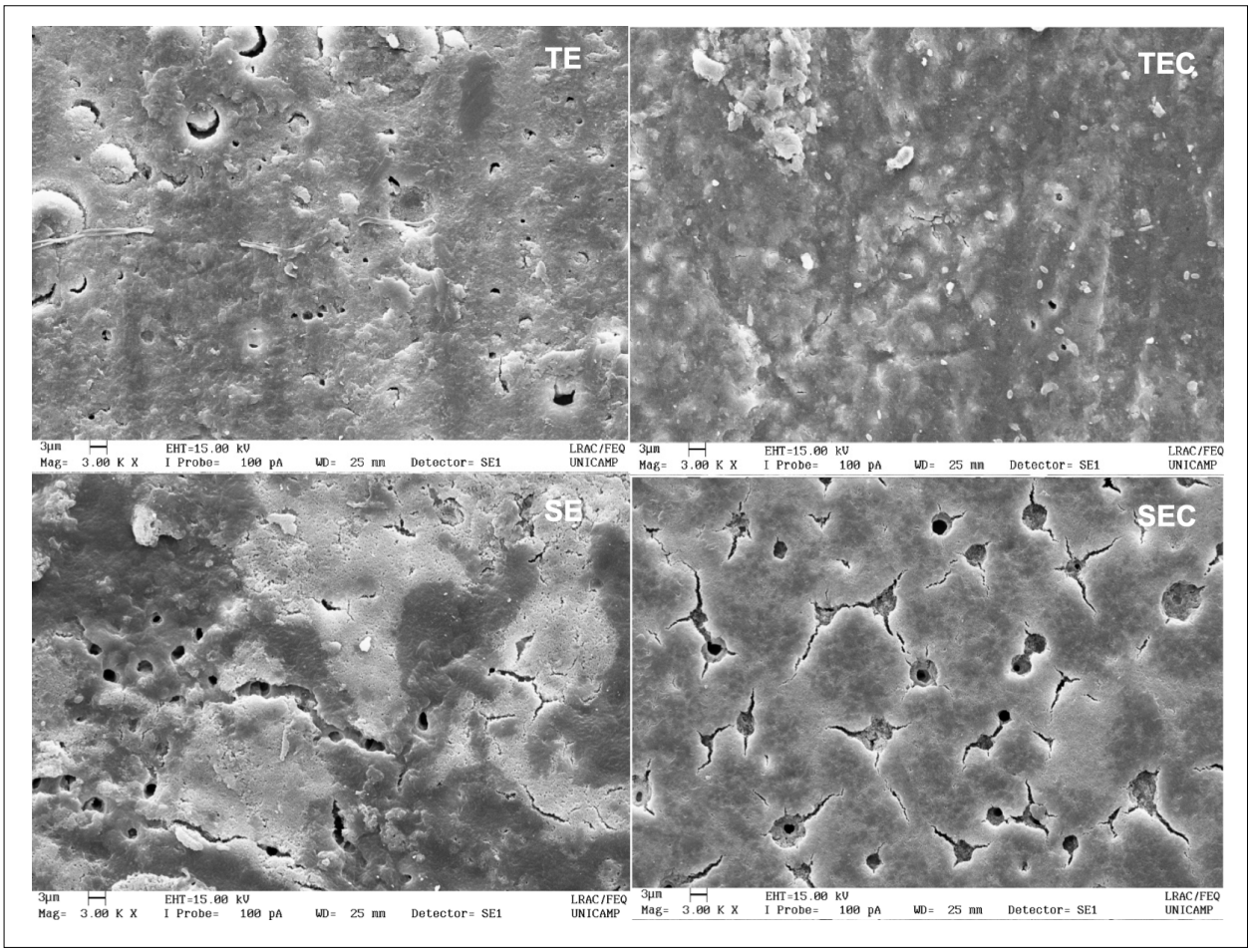

Fig. 2: *Images of dentin surface micromorphology (3000x magnification). A surface with greater uniformity and fewer pores or holes is observed with the total-etch strategy (TE and TEC). Use of the self-etching strategy (SE and SEC) caused more cracks and pores in the dentinal tubules due to greater dentin permeability. TE - Total-etch; TEC - Total-etch with chitosan; SE - Self-etching; SEC - Self-etching with chitosan.*

changes in polydispersity or particle size, but causing an impact on the colloidal stability of the solution. The higher concentration of chitosan used in the present study may therefore have influenced this stability by increasing the forces of attraction and repulsion between the particles, and causing a trend toward agglomeration. Further studies are needed to confirm this.

Chitosan may not have affected the micromorphological characteristics of the adhesive layer in the current study. Diolosà et al.[8] observed that chitosan was detected at a 100-µm depth, and explained this as a consequence of its low molecular weight (between 50 and 190 g/mol), compared to HEMA (130 g/mol) and BisGMA (512 g/mol). Dacoreggio et al.[18] observed a reduction in the number of tags in dentin, and lower bond strength, suggesting that this might be attributed to the presence of chitosan at the restorative interface. However, the higher concentration of chitosan added to the adhesive (Scotchbond Universal / 3M ESPE St. Paul, MN, USA) in the present study may be one of the factors that prevented the hydraulic conductivity of the adhesive from decreasing. This outcome, together with the characteristics of polydispersity and particle distribution, should be investigated in future studies.

In the current study, dentin permeability was higher with the total-etching bonding strategy then with the self-etching strategy, leading to the rejection of the second null hypothesis. Acid etching promotes an increase in the hydraulic conductivity of dentin[13], since the increase in the osmotic pressure exerted by the permeability measuring device promotes strong movement of dentinal fluid towards the occlusal surface,

resulting from the removal of the smear layer and smear plugs[19].

The opening of the dentinal tubules and the demineralization of the intertubular dentin allow greater penetration of the adhesive system[20], resulting in a thicker adhesive layer. The micromorphological images of the dentin section in the current study confirm the greater penetration depth of the adhesive system achieved with the total-etching strategy (≅ 10 μm). This is further corroborated by Wagner et al.[3] and Chen et al.[21] This greater penetration is attributed to the properties of the surface energy of the substrate, surface tension, and viscosity of the adhesive[22]. Moreover, the surface images show a more uniform layer with the total-etching than the self-etching strategy, which may be attributed to the thickness obtained by avoiding greater water permeation, achieved by using the adhesive system in the total-etch mode.

Maintaining the smear layer when using the self-etching strategy provides a thinner hybrid layer[3,21]. However, the reduced permeability provided by not opening the dental tubules explains the lower dentin permeability observed for this strategy in this study. The micromorphological images of the surface after use of the self-etching strategy show cracks and fissures, which may have occurred due to the higher osmotic pressure exerted on the thinner adhesive layer. Sahin et al.[19] report that adhesives containing HEMA have greater ability to absorb water which remains trapped in the resin/dentin interface.

Although universal adhesives promise easier technological processes, the benefits of the self-etching as opposed to the total-etching strategy include reducing the number of clinical steps, and lower dentin permeability. The addition of 1% chitosan did not interfere with permeability in either strategy, suggesting the need for further research into other concentrations and properties of the modified adhesive. Regarding clinical applicability, the results show that the total-etching strategy presented higher permeability than the self-etching strategy, and that the use of the self-etching strategy in Class V restorations might be better in clinical situations marked by greater hypersensitivity.

The addition of chitosan to the universal adhesive system did not affect dentin permeability. The self-etching bonding strategy led to lower dentin permeability.

DECLARATION OF CONFLICTING INTERESTS

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REFERENCES

1. Liu XX, Tenenbaum HC, Wilder RS, Quock R, Hewlett ER, Ren YF. Pathogenesis, diagnosis and management of dentin hypersensitivity: an evidence-based overview for dental practitioners. BMC Oral Health 2020 Aug 6;20(1):220. https://doi.org/10.1186/s12903-020-01199-z
2. Favaro Zeola L, Soares PV, Cunha-Cruz J. Prevalence of dentin hypersensitivity: Systematic review and meta-analysis. J Dent 2019 Feb;81:1-6. https://doi.org/10.1016/j.jdent.2018.12.015
3. Wagner A, Wendler M, Petschelt A, Belli R, Lohbauer U. Bonding performance of universal adhesives in different etching modes. J Dent 2014 Jul;42(7):800-7. https://doi.org/10.1016/j.jdent.2014.04.012
4. Fu B, Shen Y, Wang H, Hannig M. Sealing ability of dentin adhesives/desensitizer. Oper Dent 2007 Sep-Oct;32(5):496-503. https://doi.org/10.2341/06-143
5. Digole VR, Warhadpande MM, Dua P, Dakshindas D. Comparative evaluation of clinical performance of two self-etch adhesive systems with total-etch adhesive system in noncarious cervical lesions: An in vivo study. J Conserv Dent 2020 Mar-Apr;23(2):190-195. https://doi.org/10.4103/JCD.JCD_166_20
6. de Los Angeles Moyaho-Bernal M, Contreras-Bulnes R, Rodríguez-Vilchis LE, Rubio-Rosas E. Changes in deciduous and permanent dentinal tubules diameter after several conditioning protocols: In vitro study. Microsc Res Tech 2018 Aug;81(8):865-871. https://doi.org/10.1002/jemt.23048
7. Thapa B, Narain R. Mechanism, current challenges and new approaches for non-viral gene delivery. Poly Nano Gene Ther 2016; 1-27. https://doi.org/10.1016/B978-0-08-100520-0.00001-1
8. Diolosà M, Donati I, Turco G, Cadenaro M, Di Lenarda R, Breschi L, Paoletti S. Use of methacrylate-modified chitosan to increase the durability of dentine bonding systems. Biomacromolecules 2014 Dec 8;15(12):4606-13. https://doi.org/10.1021/bm5014124
9. Stenhagen ISR, Rukke HV, Dragland IS, Kopperud HM. Effect of methacrylated chitosan incorporated in experimental composite and adhesive on mechanical properties and biofilm formation. Eur J Oral Sci 2019 Feb;127(1):81-88. https://doi.org/10.1111/eos.12584
10. Ururahy MS, Curylofo-Zotti FA, Galo R, Nogueira LF, Ramos AP, Corona SA. Wettability and surface

morphology of eroded dentin treated with chitosan. Arch Oral Biol 2017 Mar;75:68-73. https://doi.org/10.1016/j.archoralbio.2016.11.017
11. Elsaka SE. Antibacterial activity and adhesive properties of a chitosan-containing dental adhesive. Quintessence Int 2012 Jul-Aug;43(7):603-13. https://doi.org/10.7555/JBR.26.20110136
12. Caldas IP, Alves GG, Barbosa IB, Scelza P, de Noronha F, Scelza MZ. In vitro cytotoxicity of dental adhesives: A systematic review. Dent Mater 2019 Feb;35(2):195-205. https://doi.org/10.1016/j.dental.2018.11.028
13. Hashimoto M, Ito S, Tay FR, Svizero NR, Sano H, Kaga M, Pashley DH. Fluid movement across the resin-dentin interface during and after bonding. J Dent Res 2004 Nov;83(11):843-8. https://doi.org/10.1177/154405910408301104
14. Ishihata H, Kanehira M, Finger WJ, Takahashi H, Tomita M, Sasaki K. Effect of two desensitizing agents on dentin permeability in vitro. J Appl Oral Sci 2017 Jan-Feb;25(1):34-41. https://doi.org/10.1590/1678-77572016-0228
15. Carvalho AO, Oliveira MT, Nikaido T, Tagami J, Giannini M. Effect of adhesive system and application strategy on reduction of dentin permeability. Braz Oral Res 2012 Sep-Oct;26(5):397-403. https://doi.org/10.1590/S1806-83242012005000020
16. Radovic I, Vulicevic ZR, García-Godoy F. Morphological evaluation of 2- and 1-step self-etching system interfaces with dentin. Oper Dent 2006 Nov-Dec;31(6):710-8. https://doi.org/10.2341/05-145
17. Oliveira TA, Anágua-Bravo E, Aoki IV, Scaramucci T, Sobral MAP. Chemical and mechanical resistance of novel experimental hybrid coatings on dentin permeability. Microsc Res Tech 2021 Feb;84(2):163-170. https://doi.org/10.1002/jemt.23574
18. Dacoreggio R, Bridi EC, Basting RT, Tenuti JGB, França FMG, Turssi CP, Amaral FLB, Basting RT. Incorporation of chitosan into a universal adhesive system: physicochemical characteristics, gelatinolytic activity, bond strength and interface micromorphology analyses. Int J Adhes Adhes 2021; 106: 102814. https://doi.org/10.1016/j.ijadhadh.2021.102814
19. Sahin C, Cehreli ZC, Yenigul M, Dayangac B. In vitro permeability of etch-and-rinse and self-etch adhesives used for immediate dentin sealing. Dent Mater J 2012;31(3):401-8. https://doi.org/10.4012/dmj.2011-217
20. Perdigão J. Current perspectives on dental adhesion: (1) Dentin adhesion - not there yet. Jpn Dent Sci Rev 2020 Nov;56(1):190-207. https://doi.org/10.1016/j.jdsr.2020.08.004
21. Chen C, Niu LN, Xie H, Zhang ZY, Zhou LQ, Jiao K, Chen JH, Pashley DH, Tay FR. Bonding of universal adhesives to dentine--Old wine in new bottles? J Dent 2015 May;43(5):525-36. https://doi.org/10.1016/j.jdent.2015.03.004
22. Sauro S, Pashley DH, Mannocci F, Tay FR, Pilecki P, Sherriff M, Watson TF. Micropermeability of current self-etching and etch-and-rinse adhesives bonded to deep dentine: a comparison study using a double-staining/confocal microscopy technique. Eur J Oral Sci 2008 Apr;116(2):184-93. https://doi.org/10.1111/j.1600-0722.2007.00518.x

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Learning strategies of dental students in Buenos Aires, Argentina prior to and during the COVID-19 pandemic

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ABSTRACT

Various theories have endeavored to explain how knowledge is accessed. Students, in order to learn, need a good repertoire of appropriate metacognitive and self-regulating strategies and knowledge, which they use consciously or unconsciously. Teachers, in addition to knowing how to teach, need to be aware of students' learning strategies, metacognition, and self-regulation, and of the impact of changes associated with recent pandemic scenarios. ***Aim:*** *The aim of this study was to identify the study strategies preferred by dental students in two different scenarios: prior to and during the pandemic.* ***Materials and Method:*** *The sample consisted of third-year dentistry students at Buenos Aires University (UBA) during 2019 (Group GP, 141 students, face-to-face activity) and 2021 (Group GE, 60 students, e-learning during the pandemic). Participants were asked (a) to provide demographic information (sex and age) and (b) to answer the abridged ACRA scale. Statistical treatment included descriptive tests; Chi², binomial exact and Student's t-test (p<0.05).* ***Results:*** *The proportion of students who participated with respect to total students enrolled was 58.50% in 2019 and 26.20% in 2021, with female gender being significantly higher. There were significant differences in total number of participants during the different periods (p=0.001), and in gender distribution during the pandemic (p=0.007). Comparison between groups GP and GE showed no significant difference regarding preferences expressed in total values for the scale or for the domains. Analysis of preferences according to gender showed significant differences in total group (p=0.007) and the domains CLCS (Cognitive and Learning Control Strategies) (p= 0.008) and LSS (Learning Support Strategies) (p=0.002). The mean values of preferences selected by females were higher. Similar results were found upon analyzing preferences during the pandemic (n= 60) considering total score (p= 0.033) and the domains CLCS (p= 0.035) and LSS (p= 0.007).* ***Conclusions:*** *The study identified trends towards an increase in the score and consequently greater use of techniques included in the domains related to metacognition, especially among women. There is potential neutralization of the impact created by the methodological shift between the two periods (face-to-face and e-learning) probably as a result of the implicit adaptability, latent in students, regarding digital methodology, which enables them to adapt to learning in challenging situations.*

Keywords*: learning strategies - e-learning - gender - COVID-19*

Impacto de la pandemia por covid-19 sobre las estrategias de aprendizaje de los estudiantes de odontología en Buenos Aires, Argentina

RESUMEN

Varias teorías se han esforzado por explicar cómo se accede al conocimiento. Para aprender, los estudiantes necesitan tener un conjunto de estrategias y conocimientos apropiados, y utilizarlos consciente o inconscientemente. Los docentes deben conocer las estrategias de aprendizaje, la metacognición y la autorregulación de los estudiantes, así como el impacto de los cambios asociados con los escenarios pandémicos recientes sobre esas estrategias. ***Objetivo:*** *El objetivo de este estudio fue identificar las estrategias de estudio preferidas por los estudiantes de odontología en diferentes escenarios: previo y durante la pandemia.* ***Materiales y Método:*** *La muestra estuvo conformada por estudiantes de tercer año de la carrera de odontología (UBA) durante el año 2019 (Grupo GP, 141 estudiantes, actividad presencial) y 2021 (Grupo GE, 60 estudiantes, e-learning durante la pandemia). Los participantes proporcionaron información demográfica (sexo y edad) y respondieron el cuestionario ACRA abreviado. El tratamiento estadístico incluyó medidas de tendencia central, dispersión y distribución de frecuencias, prueba Chi2, binomial exacta y prueba t de Student (p<0,05).* ***Resultados:*** *La distribución de estudiantes que participaron con respecto al total de estudiantes matriculados fue de 58,50% en 2019 y 26,20% en 2021, siendo significativamente mayor el género femenino. Hubo diferencias significativas en el número total de participantes durante los diferentes períodos (p=0,001) y en la distribución por género durante la pandemia (p=0,007). La comparación entre los grupos GP y GE no mostró diferencia significativa en cuanto a las preferencias expresadas en valores totales para la escala o para los dominios. El análisis de preferencias según género mostró diferencias significativas en: grupo total (p=0,007) y los dominios ECCA (Estrategias de Control Cognitivo y de Aprendizaje) (p= 0,008) y EAA (Estrategias de Apoyo al Aprendizaje) (p=0,002). Los valores medios de las preferencias seleccionadas por las mujeres fueron más altos. Resultados similares se encontraron al analizar las preferencias durante la pandemia (n= 60) considerando la puntuación total (p= 0,033) y los dominios ECCA (p= 0,035) y EAA (p= 0,007).* ***Conclusiones:*** *El estudio identificó un aumento en el puntaje y consecuentemente un mayor uso de técnicas incluidas en los dominios relacionados con la metacognición, especialmente entre las mujeres. Existe una potencial neutralización del impacto creado por el cambio metodológico entre los dos períodos (presencial y e-learning) probablemente como resultado de la adaptabilidad implícita, latente en los estudiantes, respecto a la metodología digital, que les permita adaptarse al aprendizaje en situaciones desafiantes.*

Palabras Claves*: estrategias de aprendizaje - aprendizaje virtual - género - COVID-19*

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INTRODUCTION

Various theories have endeavored to explain how knowledge is accessed. A constructivist, cyclic conception of learning[1–8] recognizes organized, coordinated mental operations or cognitive processes. These are inferred based on the subject's behavior during reasoning or problem-solving tasks which, operatively, act as the goals to be attained. This processing takes place as a path followed by the student to develop skills and learn contents: their learning method. During this process, the student employs flexible sociocultural instruments learned in contexts of interaction, applying procedures that include several specific techniques or operations in pursuit of a purpose: learning or problem-solving.

Learning to learn, or being strategic to learn, is essential in today's culture, where it is necessary to process and deal with large quantities of information and frequent challenging situations. To do so, students need to have a good repertoire of appropriate metacognitive and self-regulating strategies and knowledge, and use them consciously or unconsciously[9].

According to Díaz Barriga and Hernández Rojas[10], learning strategies are procedures or sequences of conscious, voluntary actions, and applying them involves the student knowing how to select intelligently among several resources available to him/her and knowing how to control the cognitive processes in order to complete a task successfully. This enables monitoring and assessment while a student is involved in the process according to given contextual demands and to the achievement of certain learning goals. Learning strategies are activated whenever the student is required to learn, remember or solve problems related to learning contents.

Learning strategies are propositional activities that are reflected in the four broad phases of information processing. These phases have been included in the ACRA (Acquisition, Encoding, Retrieval, Support, by its Spanish acronym) evaluation instrument[11]:

1. Information *ACQUISITION* phase, with attentional strategies (exploration and fragmentation) and repetition strategies.
2. Information *ENCODING* phase, which includes mnemotechnic, developmental and organizational strategies.
3. Information *RETRIEVAL* phase, with memory searching strategies (search for codes and clues), and answer generation strategies (planning and preparing the written answer).
4. Processing *SUPPORT* phase, which is divided into: metacognitive strategies (self-knowledge and self-management), affective strategies (self-instructions, self-control and distraction suppression), social strategies (social interactions) and motivational strategies (intrinsic motivation, extrinsic motivation and escape motivation).

Teachers, in addition to knowing how to teach, need to be aware of students' learning strategies (cognition), metacognition, and self-regulation. Activities conducted in a scenario of uncertainty created by the pandemic and the consequent methodological change (e-learning) were considered challenging. The aim of this study was therefore to identify the study strategies preferred by university students of both genders in different learning scenarios: regular and challenging.

MATERIALS AND METHOD

Sample

All third-year dentistry students during the years 2019 (GP) and 2021 (GE) were invited to take part in the study. Participation was voluntary and independent of any academic assessment. The sample was grouped as follows:

- Face-to-face group (GP), which included 141 students (58.50% of 241 total enrolled students) in the context of normal environmental situation and in-person attendance (year 2019), and
- E-learning group (GE), which included 60 students (27.39% of 219 total enrolled students) in the context of environmental challenge and e-learning (year 2021).

Students were requested (a) to provide socio-demographic information (sex and age), and (b) to answer the validated abridged ACRA scale (Acquisition, Encoding, Retrieval, Support, by its Spanish acronym)[11].

The information was requested via the same method being used for classes in each group, i.e., face-to-face for GP and virtual for GE, without teachers adding any modalization in either group.

Statistical analysis

For the descriptive analysis, the categorical variables were described by means of frequencies and percentages, and the numerical variables were

expressed as mean, minimum and maximum. Variables were compared by the Chi-square test and the binomial exact test for independent samples. Student's t-test was used for independent samples to compare the two groups with quantitative variables. Differences were considered statistically significant when p <0.05.

RESULTS

Analysis of the sample

Student participation was 58.50% of total students enrolled in the 2019 cycle (GP) and 26.20% of total students enrolled in the 2021 cycle (GE). No statistical difference was observed between the percentages of GP and GP according to gender (Chi square test: p=0.400). There were statistical differences in participation between groups GP (58.5%) and GE (26.2%) (binomial proportions test p<0.001). Thus, participation was greater in the pre-pandemic period, but the gender proportion was maintained (Table 1).

Age differed significantly between groups (pre-pandemic/post-pandemic intervention period) and gender differed significantly in the "pandemic" condition (GE) (p=0.007). Average student age was higher during the pandemic, and among females (p=0.001) (Table 2).

Analysis of scores among study strategies on the abridged ACRA scale

No statistical difference was observed between GP and GE in the total ACRA score or in the domains (Table 3).

In the whole sample (n=201), the items that significantly increased in agreement ("agree" or "strongly agree") were items 2 (p=0.030) and 3 (p=0.001) in the Cognitive Strategies and Learning Control domain. In the Learning Support Strategies Domain, item 8 showed a significant increase in disagreement ("strongly disagree") (p=0.040) (Table 4).

Table 1. Frequency distribution and percentage of students included in the study

Group	Gender		Students participating in the study	Students in the class	% Students participating
	Female	Male			
Pre-pandemic (GP) n (%)	105 (74.5%)	36 (25.5%)	141	241	58.5%
During the pandemic (GE) n (%)	48 (80.0%)	12 (20.0%)	60	229	26.2%
Total	153 (76.1%)	48 (23.9%)	201	470	42.8%

GP and GE by gender: (p=0.400)
Participation pre-pandemic and during the pandemic (0.001)

Table 2. Participating students according to age, gender and study period

Variables			Mean age ± SD (Min - Max)	N	p value
Gender		Female	22.5 ± 2.4 (19-31.0)	153	0.438º
		Male	22.2 ± 1.7 (20.0-28.0)	48	
Intervention period		GP	21.8 ± 1.9 (19.0-31.0)	141	0.001*
		GE	23.8 ± 2.5 (20.0-31.0)	60	
Intervention period	GP	Female	21.7 ± 2.0 (19.0-31.0)	105	0.321º
		Male	22.1 ± 1.8 (20.0-28.0)	36	
	GE	Female	24.1 ± 2.6 (20.0-31.0)	48	0.007*
		Male	22.6 ± 1.4 (21.0-26.0)	12	
Total participating			22.4 ± 2.3 (19.0-31.0)	**201**	

*Student's test for independent samples with Welch correction
ºStudent's test for independent samples
SD: Standard deviation
Min: minimum value
Max: maximum value

Table 3. Average scores on the ACRA scale (total score and domains), according to study period (GP and GE).

Dimension Analyzed	Period	Mean ± SD (CI95% LL - UL)	n (min -max)	p value
Total Score	Pre-pandemic	42.4 ± 3.9 (41.7-43.0)	141 (33-51)	0.904
	Pandemic	42.5 ± 4.5 (41.3-43.6)	60 (33-52)	
	Total	**42.4 ± 4.1 (41.8-43.0)**	**201 (33-52)**	
Cognitive Strategies and Learning Control	Pre-pandemic	20.0 ± 2.1 (19.7-20.4)	141 (14-24)	0.868
	Pandemic	20.1 ± 2.2 (19.5-20.6)	60 (16-24)	
	Total	**20.0 ± 2.1 (19.7-20.3)**	**201 (14-24)**	
Learning Support Strategies	Pre-pandemic	15.7 ± 2.2 (15.3-16.0)	141 (10-20)	0.479
	Pandemic	15.9 ± 2.4 (15.3-16.5)	60 (10-20)	
	Total	**15.7 ± 2.3 (15.4-16.0)**	**201 (10-20)**	
Study Habits	Pre-pandemic	6.7 ± 1.1 (6.5-6.9)	141 (4-8)	0.201
	Pandemic	6.5 ± 1.2 (6.2-6.8)	60 (3-8)	
	Total	**6.6 ± 1.2 (6.5-6.8)**	**201 (3-8)**	

Student's test for independent samples.
SD: Standard deviation
CI95%: 95% confidence interval
LL: Lower limit of the 95% confidence interval
UL: Upper limit of the 95% confidence interval

Table 4. ACRA items in which statistically significant changes are observed between study periods.

Items		Pre-pandemic	Pandemic	p value
		Frequency (%; CI95% LL-UL)	Frequency (%; CI95% LL-UL)	
Item 2	Strongly disagree	3 (2.1; 0.6 - 5.6)	0 (0.0; 0 - 0)	0.030
	Disagree	9 (6.4; 3.2 - 11.3)	1 (1.7; 0.2 - 7.5)	
	Agree	34 (24.1; 17.6 - 28.7)	24 (40.0; 32.3 - 52.6)*	
	Strongly agree	95 (67.4; 59.3 - 74.7)	35 (58.3; 45.7 - 70.2)	
Item 3	Strongly disagree	2 (1.4; 0.3 - 4.5)	0 (0.0; 0 - 0)	0.001
	Disagree	3 (2.1; 0.6 - 5.6)	4 (6.7; 2.3 - 15.1)	
	Agree	89 (63.1; 55 - 70.8)*	18 (30.0; 19.5 - 42.3)	
	Strongly agree	47 (33.3; 26 - 41.4)	38 (63.3; 50.7 - 74.7)*	
Item 8	Strongly disagree	2 (1.4; 0.3 - 2.5)	4 (6.7; 3.3 - 12.1)*	0.040
	Disagree	43 (30.5; 23.4 - 38.4)	13 (21.7; 12.7 - 33.3)	
	Agree	66 (46.8; 38.7 - 55)	23 (38.3; 26.8 - 50.9)	
	Strongly agree	30 (21.3; 15.1 - 28.6)	20 (33.3; 22.4 - 45.8)	

Chi square test, with Bonferroni post hoc
* Significant statistical difference, indicating a higher percentage between study periods.
CI95%LL: Lower limit of the 95% confidence interval
CI95%UL: Upper limit of the 95% confidence interval

- **Item 1, Cognitive Strategies and Learning Control Domain:** "In books, notes or other learning materials, I underline the words, data or phrases I consider most important in each paragraph."
- **Item 3, Cognitive Strategies and Learning Control Domain:** "I am aware of the importance of elaboration strategies, which require me to establish different kinds of associations among the contents of the study material (drawings or graphs, mental images, metaphors, self-questions, paraphrasing, etc.)."
- **Item 8, Learning Support Strategies Domain:** "I use personal resources to control my states of anxiety when they prevent me from concentrating better on studying."

Analysis of ACRA score according to gender

Analysis of the total group of students (n= 201) distributed according to gender showed significant differences (p=0.007). Analysis of domains showed significant differences between genders in the domains Cognitive and Learning Control Strategies (p= 0.008) and Learning Support Strategies (p=0.002). In all cases, females scored higher on average, expressing greater definition in the option selected for each item (Table 5). No statistical difference was observed in the Study Habits domain.

Analysis of scores during the pandemic (GE) differed significantly between genders (Table 6):

(a) Total score for the scale (p=0.033)

(b) The Control Strategies and Learning Acquisition

Table 5. Average scores on the ACRA scale (total score and domains) according to gender.

Dimension	Gender	Mean ± SD (CI95% LL - UL)	n (min -max)	p value
Total Score	Female	42.8 ± 4.1 (42.2-43.5)	153 (33-52)	0.007
	Male	41.0 ± 3.8 (39.9-42.1)	48 (33-51)	
	Total	**42.4 ± 4.1 (41.8-43.0)**	**201 (33-52)**	
Cognitive Strategies and Learning Control	Female	20.3 ± 2.2 (19.9-20.6)	153 (14-24)	0.008
	Male	19.3 ± 1.9 (18.8-19.9)	48 (16-24)	
	Total	**20.0 ± 2.1 (19.7-20.3)**	**201 (14-24)**	
Learning Support Strategies	Female	16.0 ± 2.2 (15.7-16.4)	153 (11-20)	0.002
	Male	14.8 ± 2.3 (14.2-15.5)	48 (10-19)	
	Total	**15.7 ± 2.3 (15.4-16.0)**	**201 (10-20)**	
Study Habits	Female	6.6 ± 1.2 (6.4-6.8)	153 (3-8)	0.149
	Male	6.9 ± 1.0 (6.6-7.2)	48 (4-8)	
	Total	**6.6 ± 1.2 (6.5-6.8)**	**201 (3-8)**	

Student's test for independent samples.
SD: Standard deviation
CI95%: 95% confidence interval
LL: Lower limit of the 95% confidence interval
UL: Upper limit of the 95% confidence interval
Comparison of student gender shows statistically significant differences in the total ACRA score, in Cognitive Strategies and Learning Control and in Learning Support Strategies, with females scoring higher. No statistical difference is observed in Study Habits.

Table 6. Average scores on the ACRA scale for the Total Score and in the Domains, according to gender for the pandemic group (GE)

Dimension Analyzed	Gender	Mean ± SD (CI95% LL - UL)	n (min -max)	p value
Total Score	Female	43.1 ± 4.4 (41.8-44.4)	48 (35-52)	0.033
	Male	40.0 ± 4.4 (37.2-42.8)	12 (33-48)	
	Total	**42.5 ± 4.5 (41.3-43.6)**	**60 (33-52)**	
Cognitive Strategies and Learning Control	Female	20.4 ± 2.1 (19.8-21.0)	48 (16-24)	0.035
	Male	18.9 ± 2.0 (17.7-20.2)	12 (16-23)	
	Total	**20.1 ± 2.2 (19.5-20.6)**	**60 (16-24)**	
Learning Support Strategies	Female	16.3 ± 2.3 (15.7-17.0)	48 (11-20)	0.007
	Male	14.3 ± 2.3 (12.8-15.7)	12 (10-18)	
	Total	**15.9 ± 2.4 (15.3-16.5)**	**60 (10-20)**	
Study Habits	Female	6.4 ± 1.3 (6.0-6.8)	48 (3-8)	0.279
	Male	6.8 ± 1.0 (6.2-7.5)	12 (5-8)	
	Total	**6.5 ± 1.2 (6.2-6.8)**	**60 (3-8)**	

Student's test for independent samples.
SD: Standard deviation
CI95%: 95% confidence interval
LL: Lower limit of the 95% confidence interval
UL: Upper limit of the 95% confidence interval

domain (p=0.035), and the Learning Support Strategies domain (p=0.007)

However, during the pre-pandemic period (GC), scores did not differ significantly between males and females.

DISCUSSION

Learning strategies are a construct that includes cognitive, metacognitive, motivational and behavioral elements. Based on the hypothesis that cognitive processes for information processing are acquisition, encoding or storage, and retrieval, information processing strategies can be defined as "integrated sequences of mental procedures or activities that are activated to facilitate the acquisition, storage and/or use of information". This basic hypothesis is included in the theories about mental representation of knowledge of memory and in the "instructional" approach[12]. These theories hypothesize that the brain operates as if it were the outcome of three basic cognitive processes: a) acquisition, b) encoding or storage, and c) retrieval or evocation. In addition, other metacognitive, affective and social processes are needed, which are addressed by support strategies. Mental procedures or management strategies, called "micro strategies, learning tactics or study strategies", can be deduced from knowledge of the cognitive processes. The ACRA scale was developed based on this theoretical framework[13].

Today, distance learning and particularly the use of Information and Communication Technologies in the educational process have revealed the need to develop attitudes of autonomy, self-direction and self-regulation in the learning process by promoting **strategic learning**, where mental representation (learning) is related to relevance to everyday use and significance to the learner's context.

The results of the current study reveal moderate interest in participating among students in the GP group, and lower interest among students in the GE group, with significant differences between participation proportions (p=0.001). This may be interpreted as one of the expressions of consequences of the social impact of the pandemic challenge. These results were similar to those reported by Turkyilmaz et al.[14] in a study in which 22.6% of 1130 pre-doctoral students responded. Other studies achieved more representative responses[15].

Different aspects of the nature of the social impact caused by the pandemic were reported by Bhattacharya[16]. From a medical point of view, several examples showed the impact of the pandemic. Gondolesi et al.[17] showed a 55% reduction in liver transplants, mainly as a consequence of the high level of occupancy of hospital beds by COVID-19 patients. Navarro Rubio et al.[18] highlighted the impact of the pandemic on the level of healthcare and in other spheres of society, including response to the demands of the patients involved, their families and legal representatives.

Ali and Alharbi[19] say that there is an urgent need to educate the new generation in science and technology so that they are prepared to react to any disaster of this kind in the future. They recommend providing training for prevention and adequate management of essential resources for combatting COVID-19 or other potential risks. It may be conjectured that, to encourage students to respond to research for generating scientific evidence (SE), integrated validated instruments should be used, and training workshops should be provided to ensure competence in distinguishing between scientific evidence-based and non-evidence-based studies. Such competence would complement skills related to handling mobile technology, which young students usually have[20].

Analysis of the differences between genders

Strategic learning is promoted through self-regulation techniques and the adoption of those that adapt to (a) the student's learning style, (b) the student's context, (c) the requirements of each subject, and (d) the creation of the most appropriate learning setting to contribute to the efficacy of the process and the acquisition of general and specific competencies.

Although the differences recorded in the current study did not have an impact when the total population was analyzed, tendencies were found of greater preference by females to adopt learning strategies tending to metacognition in the LSS domain.

The preferences recognized in the answers to the ACRA scale may reflect greater commitment to cognitive and metacognitive strategies because they enable maintenance of an appropriate mental state for learning. They include strategies to foster motivation and concentration, reduce anxiety, focus attention on the task at hand and organize study time, among others.

Learning support strategies have indirect impact on the information to be learned, and their purpose is to improve the level of cognitive functioning. The differences found in the increase in strategies by females for the two periods (p=0.007) included in the current study have also been reported by other authors, mainly in the CSLC and LSS domains. Faria and Montaine[21] reported an interaction effect between gender and study outcomes over the course of academic degree studies, showing the existence of dynamic, effort-dependent conceptions; persistence, and other intelligence variables in women, while conceptions were more static in men. Studies on different academic performance have established that women students have better levels of academic motivation, especially during stages immediately prior to access to university.

In the university setting, consistent results have been found regarding the higher number of strategies used by female students. García-Garcia et al.[22] reported that most of the female students analyzed used more and better strategies for acquisition, retrieval and processing support, as well as encoding strategies, with greater use of information organizational techniques. Overall, these authors found that a higher percentage of female students used implicit strategies in the different processing phases, as a result of their prior experience. Among the supporting strategies, they highlight processing, higher level of metacognitive techniques in all senses (cognitive awareness and strategy adjustment), as well as better study planning and use of motivational-affective techniques. Rogers et al.[23] showed the differences regarding student motivational style based on sex, although depending on the type of learning, as was confirmed in our study by the significant differences found in females regarding preferences for item 8**,** specifically linked to motivational aspects.

Women students have been reported to make greater use of regulatory learning strategies[22] and study organization and planning strategies. These results are confirmed in the current study by the significant differences recorded for female students during the pandemic for items 9 and 11 of domain LSS[24].

The increase in certain records of the strategies and techniques identified as instrumental to significant learning acquisition and favorable to metacognition are unknown factors in the evidence in the field of education. More in-depth knowledge of these strategies, complemented by neuroscience studies, should be considered when designing teaching strategies. Based on Gardner's theory of multiple intelligences, Collins[25] claims that different parts of the brain are responsible for competencies that everyone possesses to some degree. These multiple intelligences could be used as strategies to improve learning.

A recent study on animals by Chen et al.[26] found that sex is a proxy for multiple genetic and endocrine influences on behavior, including how environments are sampled. Differences have also been analyzed according to gender in student and graduate perception regarding visual, aural, read/write and kinesthetic (VARK) learning styles and outcomes in examinations, with significant association between perceived VARK mode and outcomes in examinations[27–29].

Analysis of learning methods and techniques

It is important to use methods to complement expository teaching methods, such as problem-solving, case studies, use of questions, class discussion, projects, cooperative work, student participation and commitment, and to use formative assessment methods to complement summative assessment and provide feedback to the student[30]. Choe et al.[31] reported a high degree of satisfaction when Mayer's principles of multimedia learning were applied.

Turkyilmaz et al.[14] assessed the influence of e-learning on dental education as perceived by predoctoral dental students. That study found that the most important factor for online applications influencing academic performance was the "**organization and logic of content**" (54%). Their results indicated that e-learning may successfully be used in a dental school curriculum to enhance students' perceptions of fundamental concepts, and to enable students to apply this knowledge to clinical cases. The outcomes seem to agree with our study, which was conducted on mid-level dentistry students. It is essential to conduct further research on mid-level dental students' preferences regarding social networks, online applications and databases, in order to include e-learning in course subjects. In our study, during the pandemic period, when students increased their use of e-learning strategies, females scored significantly higher than males in the CSLC and LSS domains.

Abbasi et al.[15] analyzed perception of and satisfaction

with the use of e-learning during the pandemic in 11 countries with different development categories, revealing preference for the use of Zoom (41%), and reporting interference in e-learning due to problems with Internet. Most participants agreed that e-learning was satisfactory for acquiring knowledge, but not effective for acquiring clinical and technical skills.

As the COVID-19 lockdown eases, there is a need to improve e-learning methods. Blended experiences are recommended for students in the field of healthcare. Some alternatives are the development of problem-based learning[32], challenge-based learning, or disruptive methods such as gamification. Seidlein et al.[33] claim that complementary gamified e-learning tools are promising, considering the different levels of knowledge among students and the changing behavior of learning. Turner et al.[34] stated that the differences in answers between millennial students and their teachers need to be overcome regarding the use of case studies, study guides and group work. Interdisciplinary work combining social psychology and cognitive neuroscience would enable understanding in terms of interactions at social, cognitive and neuronal levels.

CONCLUSIONS

The significant differences in the preferences expressed by students and identified in the domains and items of the abridged ACRA scale during the different learning periods do not show significant impact on the total score for the abridged ACRA scale. The study identified trends towards an increase in the score and consequently greater use of techniques included in the domains related to metacognition, especially among women. It may be conjectured that there is potential neutralization of the impact created by the methodological shift between the two periods (face-to-face and e-learning) as a result of the implicit adaptability, latent in students, regarding digital methodology, which enables them to adapt to learning in challenging situations, as shown by the increase in some of the items in domains CLCS and LSS.

DECLARATION OF CONFLICTING INTERESTS

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Bruner J. Toward a Theory of Instruction. Belkapp Press; 1966.
2. Ausubel D. Educational Psychology. A Cognitive View. Holt, Rinehart and Winston, Inc; 1968.
3. Ausubel DP. The Nature of Meaning and Meaningful Learning BT - The Acquisition and Retention of Knowledge: A Cognitive View. In: Ausubel DP, ed. Springer Netherlands; 2000:67-100. https://doi.org/10.1007/978-94-015-9454-7
4. Gagné RM. The Conditions of Learning and Theory of Instruction. Holt, Rinehart and Winston, Inc., Florida-ABD, 4th edition, 1985.
5. Flavell JH. Cognitive Development. 4th Edition. 1977. Englewood Cliffs, NJ: Prentice-Hall.
6. Bandura A. Social Foundations of Thought and Action: A Social Cognitive Theory. Prentice Hall; 1986.
7. Feuerstein R, Hoffman M, Miller R. Instrumental Enrichment: An Intervention Program for Cognitive Modifiability. University Park Press; 1980.
8. Sternberg RJ. The Theory of Successful Intelligence. *Rev Gen Psychol*. 1999;3(4):292-316. https://doi.org/10.1037/1089-2680.3.4.292
9. Pozo J, Monereo C, Castelló M. El uso estrategico del conocimiento. In: Alianza Editorial; 2001:211-234.
10. Diaz Barriga F, Hernandez Rojas G. Estrategias docentes para un aprendizaje significativo, una interpretacion cognitiva. 2nd edition. Mc Graw Hill; 2005. http://creson.edu.mx/Bibliografia/Licenciatura en Educacion Primaria/Repositorio Planeacion educativa/diaz-barriga---estrategias-docentes-para-un-aprendizaje-significativo.pdf
11. De la Fuente Arias J, Justicia Justicia F. Escala de estrategias de aprendizaje ACRA-Abreviada para alumnos universitarios. Electron J Res Educ Psychol. 2017;1(2). https://doi.org/10.25115/ejrep.2.114
12. Bernad Mainar J. Modelo cognitivo de evaluación educativa. escala de estrategias de aprendizaje contextualizado. 1st. edition. Madrid, España, Narcea Ediciones; 2000.
13. Román J, Gallego S. Escalas de Estrategias de Aprendizaje. Tea Ediciones; 2008. http://www.web.teaediciones.com/Ejemplos/ACRA_extracto_web.pdf
14. Turkyilmaz I, Hariri NH, Jahangiri L. Student's Perception of the Impact of E-learning on Dental Education. J Contemp Dent Pract. 2019;20(5):616-621. https://doi.org/10.5005/jp-journals-10024-2568
15. Abbasi MS, Ahmed N, Sajjad B, incluir todos los autores. E-Learning perception and satisfaction among health sciences students amid the COVID-19 pandemic. Work. 2020;67(3):549-556. https://doi.org/10.3233/WOR-203308

16. Bhatticharya S. The Social Impact of the COVID-19 Pandemic. Observer Research Foundation. 2020;(406):14. https://policycommons.net/artifacts/1424379/the-social-impact-of-the-covid19-pandemic/2038653/
17. Gondolesi GE, Reyes-Toso ML, Bisigniano L. Impact of covid-19 pandemic in liver transplantation in Argentina. Other collateral damage. Medicina (B Aires). 2020;80(6):71-82.
18. Navarro Rubio MD, Baquero Úbeda JL, Bosque García AM, Alfonso Zamora S, Lorenzo Garmendia A. Impact of the COVID-19 pandemic: The point of view of patient associations TT - Impacto de la pandemia por COVID-19: El punto de vista de las asociaciones de pacientes. J Healthc Qual Res. 2021;36(6):355-362. https://doi.org/10.1016/j.jhqr.2021.05.003
19. Ali I, Alharbi OML. COVID-19: Disease, management, treatment, and social impact. Sci Total Environ. 2020;728: 138861. https://doi.org/10.1016/j.scitotenv.2020.138861
20. Khatoon B, Hill K, Walmsley AD. Mobile learning in dentistry: challenges and opportunities. *Br Dent J.* 2019;227(4):298-304. https://doi.org/10.1038/s41415-019-0615-x
21. Faria L, Fontaine AM. Adolescents' personal conceptions of intelligence: The development of a new scale and some exploratory evidence. Eur J Psychol Educ. 1997;12:51-62. https://doi.org/10.1007/BF03172869
22. García García L, Hernández Hernández P, Henríquez I. Estrategias cognitivas y motivacionales según carreras universitarias. Evaluación e Interv Psicoeduc Rev iInteruniversitaria Psicol la Educ. 1998;1:35-52.
23. Rogers CG, Galloway D, Armstrong D, Leo E. Gender differences in motivational style: a comparison of measures and curriculum area. Br J Educ Psychol. 1998;68 (Pt 2):189-202. https://doi.org/10.1111/j.2044-8279.1998.tb01283.x
24. De la Fuente Arias J, Justicia Justicia F. Diferencias de género en las técnicas de aprendizaje de los alumnos universitarios. Rev galego-portuguesa psicoloxía e Educ. 2001;7:239-248.
25. Collins JW. The Neuroscience of Learning. J Neurosci Nurs. 2007;39(5). https://doi.org/10.1097/01376517-200710000-00008
26. Chen CS, Ebitz RB, Bindas SR, Redish AD, Hayden BY, Grissom NM. Divergent Strategies for Learning in Males and Females. Curr Biol. 2021;31(1):39-50.e4. https://doi.org/10.1016/j.cub.2020.09.075
27. Dobson JL. A comparison between learning style preferences and sex, status, and course performance. Adv Physiol Educ. 2010;34(4):197-204. https://doi.org/10.1152/advan.00078.2010
28. Liew S-C, Sidhu J, Barua A. The relationship between learning preferences (styles and approaches) and learning outcomes among pre-clinical undergraduate medical students. BMC Med Educ. 2015;15:44. https://doi.org/10.1186/s12909-015-0327-0
29. Murphy RJ, Gray SA, Straja SR, Bogert MC. Student learning preferences and teaching implications. J Dent Educ. 2004;68(8):859-866. https://doi.org/10.1002/j.0022-0337.2004.68.8.tb03835.x
30. Smith KA, Douglas TC, Cox MF. Supportive teaching and learning strategies in STEM education. New Dir Teach Learn. 2009;(117):19-32. https://doi.org/10.1002/tl.341
31. Choe RC, Scuric Z, Eshkol E, et al. Student Satisfaction and Learning Outcomes in Asynchronous Online Lecture Videos. CBE Life Sci Educ. 2019;18(4):ar55. https://doi.org/10.1187/cbe.18-08-0171
32. Holen A, Manandhar K, Pant DS, et al. Medical students' preferences for problem-based learning in relation to culture and personality: a multicultural study. Int J Med Educ. 2015;6:84-92. https://doi.org/10.5116/ijme.558e.6451
33. Seidlein A-H, Bettin H, Franikowski P, Salloch S. Gamified E-learning in medical terminology: the TERMInator tool. BMC Med Educ. 2020;20(1):284. https://doi.org/10.1186/s12909-020-02204-3
34. Turner AM, Prihoda TJ, English DK, Chismark A, Jacks ME. Millennial Dental Hygiene Students' Learning Preferences Compared to Non-Millennial Faculty Members' Teaching Methods: A National Study. J Dent Educ. 2016;80(9):1082-1090. https://doi.org/10.1002/j.0022-0337.2016.80.9.tb06190.x

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Dental students' satisfaction with their course and how it is associated to their satisfaction with life and career outlook

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ABSTRACT

Identifying factors that influence the satisfaction of undergraduate dental students can contribute to establishing strategies for improving the educational process, furthering student development, and enhancing students' academic and professional achievements. ***Aim:*** *the present study investigated associations between undergraduate students' satisfaction with the dental course and their satisfaction with life, perception of professional career and sociodemographic factors.* ***Materials and method:*** *this was a cross-sectional study on 512 dental students from a Brazilian School of Dentistry. Students were asked to respond to three questionnaires about sociodemographic characteristics, academic, and professional perceptions. Satisfaction with life was measured using the Brazilian version of the Satisfaction with Life Scale, which comprises five statements with responses scored on a seven-point Likert scale ranging from "I strongly disagree" to "I strongly agree". Unadjusted and adjusted multiple logistic regression analyses were performed to test associations between satisfaction with the dental course and key variables.* ***Results:*** *most participants were female (73.2%), single (99.8%) and with mean age 21.77 (±2.71) years. Approximately three quarters had a family income over three times higher than the national monthly minimum wage. Higher satisfaction with the course was associated with more optimistic perception of the job market (OR=2.44; 95% CI: 1.36-4.40), better academic performance (OR=1.65; 95% CI: 1.00-2.74) and greater satisfaction with life (OR=1.10; 95% CI: 1.06-1.15). Students' sex and family income did not have significant impact on satisfaction with the dental course.* ***Conclusions:*** *satisfaction with the dental course is related to an optimistic perception of the job market, academic performance and satisfaction with life.*

Keywords: *dental education - dental student - professional education - personal satisfaction - academic success*

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Satisfação com o curso de Odontologia e suas associações com a satisfação com a vida e perspectiva de carreira

RESUMEN

Identificar os fatores que influenciam na satisfação dos estudantes de odontologia pode contribuir para o estabelecimento de estratégias que aprimorem o processo educacional, favorecendo o desenvolvimento dos alunos e potencializando suas conquistas acadêmicas e profissionais. ***Objetivo:*** *o presente estudo investigou as associações entre a satisfação de estudantes de graduação com o curso de odontologia e sua satisfação com a vida, percepção da carreira profissional e fatores sociodemográficos.* ***Materiais e método:*** *um estudo transversal foi realizado com 512 estudantes de Odontologia de uma Faculdade Brasileira de Odontologia. Três questionários foram aplicados aos alunos, com o objetivo de avaliar as características sociodemográficas, percepções acadêmicas e profissionais. A satisfação com a vida foi mensurada pela versão brasileira da Escala de Satisfação com a Vida, que compreende cinco afirmações com respostas pontuadas em uma escala Likert de sete pontos, variando de "discordo totalmente" a "concordo totalmente". A análise de regressão logística foi realizada para testar associações entre a satisfação com o curso de Odontologia e as demais variáveis. Análises de regressão logística múltipla não-ajustada e ajustada foram realizadas para testar associações entre a satisfação com o curso de odontologia e variáveis-chave.* ***Resultados:*** *A maioria dos participantes era do sexo feminino (73,2%), solteiros (99,8%) e com média de idade de 21,77 (±2,71) anos. Aproximadamente três quartos tinham renda familiar três vezes superior ao salário-mínimo nacional. A maior satisfação dos alunos com o curso foi associada a maior percepção otimista do mercado de trabalho (OR=2,44; IC 95%: 1,36-4,40), desempenho acadêmico (OR=1,65; IC 95%: 1,00-2,74) e satisfação com a vida (OR =1,10; IC 95%: 1,06-1,15). O sexo dos alunos e a renda familiar não apresentaram impacto significativo na satisfação com o curso de odontologia.* ***Conclusão:*** *a satisfação com o curso de odontologia está relacionada com a percepção otimista do mercado de trabalho, desempenho acadêmico e satisfação com a vida.*

Palavras-Chave: *educação odontológica - estudante de odontologia - educação profissional - satisfação pessoal - sucesso acadêmico*

INTRODUCTION

Emotional and social aspects are vital to the development and maintenance of interpersonal relations, including those between healthcare providers and patients[1]. The emotional reactions of university students are reflected by their academic satisfaction[2]. Students' satisfaction with their undergraduate course may involve factors such as academic performance[4,5], motivation in the choice of course[4] and job prospects[6]. Dissatisfaction can affect their university life and learning processes, and compromise their professional future, work environment and relationships with other healthcare providers. Even so, psychometric tests are rarely administered in health courses at universities[3].

The literature reports associations between dental students' satisfaction with their undergraduate course, sociodemographic characteristics[5,7], and satisfaction with life[8,9], though there are few studies on the subject[4,6,7].

Knowledge of factors that influence degree of satisfaction among undergraduate dental students can contribute to establishing strategies for improving the educational process, furthering student development, and enhancing academic and professional achievements. Dental teaching institutions could use such knowledge to take specific actions to improve their academic role.

The aim of this study was thus to investigate associations between undergraduate dental students' satisfaction with their course and its association with their demographic characteristics, satisfaction with life, and other professional factors, in a representative sample from a public university in Brazil.

MATERIALS AND METHOD

Ethical aspects

This study was approved by the Human Research Ethics Committee of *Universidade Federal de Minas Gerais* (UFMG), Brazil (protocol number: 67189617.2.1001.5149). All volunteers received explanations regarding the objectives of the study and agreed to participate by signing a statement of informed consent.

Study design and sample characteristics

This was a cross-sectional study on dental students from the School of Dentistry of the UFMG, which is the fifth largest university in Latin America and the third largest higher education institution in Brazil, according to Times Higher Education ranking published in July 2021[10].

The inclusion criterion was being an undergraduate student enrolled in the UFMG dental course. The universe population comprised 641 undergraduate dental students, of whom 512 took part in the study (response rate: 80.37%), as 129 were not present at the time of data collection.

Data collection

A pilot study was first conducted with ten students to assess the logistics of administering the questionnaire and participant adherence. The results of the pilot study did not show any need for changes in the proposed study method.

Data were collected in the second semester of 2018. Self-administered structured questionnaires were used, addressing sociodemographic characteristics, the dental course, professional career, and satisfaction with life. Sociodemographic variables were age, sex, and income. Income was dichotomized as < three times or ≥ three times the Brazilian monthly minimum wage (R$ 954/US$ 170). Questions on the course and professional career enquired about whether students had jobs while they were in the undergraduate course, and their degree of satisfaction with the course, course choice process, academic performance, and perceptions regarding the job market. The Satisfaction with Life Scale (SWLS) was used, which consists of five statements scored on a seven-point Likert scale with response options ranging from "I strongly disagree" (1 point) to "I strongly agree" (7 points). Higher scores indicate greater satisfaction with life. Cronbach's alpha coefficient for the present sample was $\alpha = 0.82$[11].

Data analysis

The *Statistical Package for the Social Sciences* (SPSS for Windows, version 21.0, IBM Inc., Armonk, NY, USA) was used for data organization and statistical analysis. Data frequency distribution was determined for sample characterization. Logistic regression analysis was performed to test associations between the dependent variable satisfaction with the dental course and the independent variables. The unadjusted *backward stepwise* procedure was used for selecting variables with a p-value <0.20 to be used in the adjusted analyses.

RESULTS

Table 1 shows participants' characteristics. Most participants were female (73.2%) and single (99.8%). Monthly family income was higher than US$ 1,755.82 among 77.4% of the sample. Mean age was 21.77 ± 2.71 years and most students expressed high satisfaction with the course (80.1%). For 51.8%, dentistry was not their first course of choice. Among these, medicine had been the first educational and professional choice for 72.5%. Regarding satisfaction with life, the mean SWLS score was 24.13 ± 6.47 points (range: 5 to 35 points). Table 2 shows the results of the logistic regression analysis conducted to determine factors associated with the outcome. The multivariate analysis revealed that satisfaction with the course was associated with perception of the job market (OR = 2.44; 95% CI: 1.36-4.40), academic performance (OR = 1.65; 95% CI: 1.00-2.74) and satisfaction with life (OR = 1.10; 95% CI: 1.06-1.15).

Table 1. Characterization of sample (n = 512).

Variables	Frequency	
	N	%
Sex		
Male	137	26.8
Female	375	73.2
Family income		
< 3 x monthly min. wage (R$ 954/ U$ 170)	116	22.6
≥ 3 x monthly min. wage (R$ 954/ U$ 170)	396	77.4
Jobs		
Not employed	476	93.0
Employed	36	7.0
Dentistry as first choice of course		
No	265	51.8
Yes	247	48.2
Performance in course		
Low	267	52.1
High	245	47.9
Perceived prospects in job market		
Low	323	63.0
High	189	37.0
Satisfaction with dental course		
Low	102	19.9
High	410	80.1

***Age:** mean and standard deviation: 21.77 (± 2.71)
***Satisfaction with life:** mean and standard deviation: 24.13 (± 6.47)

DISCUSSION

Personal, academic, professional, and sociodemographic factors may be related to satisfaction with the course[4-9]. In the present study, satisfaction with the course was related to perception of the job market, academic performance and satisfaction with life, but not to sociodemographic variables. The association between perception of the job market and satisfaction with the dental course is related to optimistic outlooks. Optimistic students express considerable interest in their professional future, are enthusiastic about the field of learning, feel confident about succeeding in their careers, and express high levels of satisfaction with the course[7]. Self-efficacy is associated with higher likelihood of students persevering to complete their studies and having greater confidence regarding success in the world of work. Programs that promote self-esteem and confidence among dental students regarding a professional future should therefore be encouraged[12,13]. Students' self-efficacy can be fostered by providing training to improve self-awareness for increasing knowledge and skills related to the profession. The literature reports that enhanced self-efficacy generates self-determined motivation and greater satisfaction with the course. Academic institutions can help motivate students regarding their professional future by promoting satisfaction with the course and life itself[13].

The study also found an association between academic performance and satisfaction with the course. Highly motivated dental students tend to have better learning characteristics[14]. Educational motivation involves engagement for pleasure and satisfaction[5], and satisfied students tend to be motivated, resulting in a good academic performance. Studies on students of medicine and psychology in Korea and Australia also found an association between satisfaction and academic performance[15,16]. However, a study on undergraduate students of social work in Germany found no association between satisfaction and academic performance[17]. Satisfaction with the course may be related differently to academic performance in different professional fields such as health and human sciences.

Satisfaction with life was also associated with satisfaction with the dental course. Satisfaction with life is not the mere sum of satisfaction in different dimensions of life, but rather, a stable psychological characteristic that influences the perceptions and

Table 2. Logistic regression for association between satisfaction with dental course and independent variables (n = 512).

Variables	Satisfaction with dental course		Unadjusted OR (95% CI)	p-value	Adjusted OR (95% CI)	p-value
	Low N (%)	High N (%)				
Sex						
Male	29 (21.2)	108 (78.8)	1.00		-	-
Female	73 (19.5)	302 (80.5)	1.11 (0.69-1.80)	0.670	-	-
Family income						
< 3 x monthly min. wage (R$ 954/ U$ 170)	24 (20.7)	92 (79.3)	1.00		-	-
≥ 3 x monthly min. wage (R$ 954/ U$ 170)	77 (19.4)	319 (80.6)	1.10 (0.66-1.84)	0.717	-	-
Jobs						
Not employed	95 (20.0)	381 (80.0)	1.00		-	-
Employed	7 (19.4)	29 (80.6)	1.03 (0.44-2.43)	0.941	-	-
Dentistry as first choice of course						
No	65 (24.5)	200 (75.5)	1.00		1.00	
Yes	34 (13.8)	213 (86.2)	1.99 (1.25-3.17)	**0.004**	1.62 (0.98-2.67)	0.059
Performance in course						
Low	71 (26.6)	196 (76.4)	1.00		1.00	
High	31 (12.7)	214 (87.3)	2.5 (1.57-.98)	**<0.001**	1.65 (1.00-2.74)	**0.052**
Perceived prospects in job market						
Low	83 (25.7)	240 (74.3)	1.00		1.00	
High	19 (10.1)	170 (89.9)	3.11 (1.82-5.31)	**<0.001**	2.44 (1.36-4.40)	**0.003**
Satisfaction with life	-	-	1.13 (1.09-1.17)	**<0.001**	1.10 (1.06-1.15)	**<0.001**

reactions of individuals in their environment[9,18,19]. The concepts of happiness and satisfaction with life are similar, and their assessment is subjective, depending on self-perception, mood and lifestyle[20]. Despite cultural differences, other studies on students in Hong Kong and Mexico found the same association as the current study, with people who were more satisfied with life reporting greater satisfaction with the course[9,21].

Students in Brazil are admitted to public universities based on their grades in a national admission examination which evaluates the knowledge acquired at high school. Twice a year, an online system is opened to rank the exam grades, after which students can apply for up to two courses anywhere in the country. The higher the grade, the greater the possibility of being admitted[22]. Admission to some undergraduate courses such as medicine can be difficult as a result of the high level of competition among candidates[23]. Students who are not admitted to the medical course often choose dentistry as a second option. This might cause frustration, as dentistry may not have been chosen based on vocation[24]. In the present study, half the students reported that dentistry was not their course of choice. However, this finding was not related to satisfaction with the course, possibly because medicine and dentistry are both in the health field and share subjects and methods, or because students who did not identify with dentistry dropped out of the course in the initial semesters.

None of the sociodemographic factors studied (age, sex or having a job) influence students' satisfaction with the course. However, previous studies using the Academic Motivation Scale found that women had greater self-determination than men and greater motivation regarding the dental course[5,25]. A study conducted with computer science students at a public university in Spain found that women tended to express greater satisfaction with the course than men[8]. Sociodemographic indicators should be explored in future studies to enable a more in-depth understanding of these divergent results.

The present study has strengths and limitations. The

high response rate ensured good representativeness of the target population. However, the cross-sectional design does not enable the establishment of cause-and-effect relationships. Longitudinal studies are needed to identify causality among the study variables and contribute more information upon which to base intervention measures.

Teaching institutions should develop programs to foster students' self-awareness, empowerment, and mental and emotional healthcare. Curricular opportunities should be created to promote undergraduate dental students' self-reflection and self-efficacy, and help improve their attitude towards the job market by pointing out both challenges and potential victories. This would help keep students motivated, optimistic and satisfied with life.

CONCLUSION

Dental undergraduate students who were more optimistic about their prospects in the job market, those with a high academic performance, and those satisfied with life were more satisfied with their dental course. Age, sex, and income were not associated with such satisfaction.

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DECLARATION OF CONFLICTING INTERESTS

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Azimi S, AsgharNejad Farid AA, Kharazi Fard MJ, Khoei N. Emotional intelligence of dental students and patient satisfaction. Eur J Dent Educ. 2010;14(3):129-132. https://doi.org/10.1111/j.1600-0579.2009.00596.x
2. Al-Eyadhy A, Alenezi S. The impact of external academic accreditation of undergraduate medical program on students' satisfaction. BMC Med Educ. 2021; 21(1):565. https://doi.org/10.1186/s12909-021-03003-0
3. Birks Y, McKendree J, Watt I. Emotional intelligence and perceived stress in healthcare students: a multi-institutional, multi-professional survey. BMC Med Educ. 2009; 9:61. https://doi.org/10.1186/1472-6920-9-61
4. Wach F, Karbach J, Ruffing S, Spinath FM. University students' satisfaction with their academic studies: Personality and motivation matter. Front Psychol. 2016;7: 55. https://doi.org/10.3389/fpsyg.2016.00055
5. Orsini C, Binnie VI, Wilson SL. Determinants and outcomes of motivation in health professions education: a systematic review based on self-determination theory. J Educ Eval Health Prof. 2016;13:19. https://doi.org/10.3352/jeehp.2016.13.19
6. McIlveen P, Beccaria G, Burton LJ. Beyond conscientiousness: Career optimism and satisfaction with academic major. J Vocat Beh. 2013;83(3):229-236. https://doi.org/10.1016/j.jvb.2013.05.005
7. Letcher W, Neves JS. Determinants of undergraduate business student satisfaction. Res High Educ. 2010;6:1. http://www.aabri.com/manuscripts/09391.pdf
8. Moro-Egido AI, Panades J. An analysis of student satisfaction: Full-time vs. part-time students. Soc Indic Res. 2010;96(2):363-378. https://doi.org/10.1007/s11205-009-9482-1
9. Shek DTL, Chai W. The impact of Positive Youth Development Attributes and Life Satisfaction on academic well-being: A longitudinal mediation study. Front Psychol. 2020;11:2126. https://doi.org/10.3389/fpsyg.2020.02126
10. Times Higher Education. Word University Rankings**.** 2021. https://www.timeshighereducation.com/content/world-university-rankings
11. Zanon C, Bardagi MP, Layous K, Hutz CS. Validation of the Satisfaction with Life Scale to Brazilians: Evidences of measurement noninvariance across Brazil and US. Soc Indic Res. 2014;119(1):443–453. https://doi.org/10.1007/s11205-013-0478-5
12. Aditya S. Motivations and future aspirations of dental interns: a cross-sectional study. SRM J Res Den Sci. 2013;4(3):114-118. https://doi.org/10.4103/0976-433X.121635
13. Komarraju M, Swanson J, Nadler D. Increased career self-efficacy predicts college students' motivation, and course and major satisfaction. J Career Assess. 2014;22(3):420-432. https://psycnet.apa.org/doi/10.1177/1069072713498484
14. Orsini CA, Binnie VI, Tricio JA. Motivational profiles and their relationships with basic psychological needs, academic performance, study strategies, self-esteem, and vitality in dental students in Chile. J Educ Eval Health Prof. 2018;19;15:11. https://doi.org/10.3352/jeehp.2018.15.11
15. Park J, Chung S, An H, Park S et al. A structural model of stress, motivation, and academic performance in medical students. Psychiatry Investig. 2012;9:143-49. https://doi.org/10.4306/pi.2012.9.2.143
16. Bailey TH, Phillips LJ. The influence of motivation and adaptation on students' subjective well-being, meaning in life and academic performance. High Educ Res

Dev. 2016;35:201–16. https://doi.org/10.1080/07294360.2015.1087474
17. Blanz M. How do study satisfaction and academic performance interrelate? An investigation with students of Social Work programs. Eur J Soc Work. 2014;17(2):281-292. http://dx.doi.org/10.1080/13691457.2013.784190
18. Diener E. Subjective well-being. Psychol Bull. 1984;95(3):542–575. https://psycnet.apa.org/doi/10.1037/0033-2909.95.3.542
19. Schimmack U, Diener E, Oishi S. Life-satisfaction is a momentary judgment and a stable personality characteristic: the use of chronically accessible and stable sources. J Pers. 2002;70(3):345-384. https://doi.org/10.1111/1467-6494.05008
20. Ortiz FR, Paiva SM, Pordeus IA, Ardenghi TM. Psychometric properties and longitudinal measurement invariance of the Brazilian version of the subjective happiness scale in adolescents. J Clin Transl Res. 2021;7(2):234-240. http://www.ncbi.nlm.nih.gov/pmc/articles/pmc8177854/
21. Ojeda L, Flores LY, Navarro RL. Social cognitive predictors of Mexican American college students' academic and life satisfaction. J Couns Psychol. 2011;58(1):61-71. https://doi.org/10.1037/a0021687
22. Portal Único de Acesso ao Ensino Superior. Ministério da Educação. https://acessounico.mec.gov.br/sisu.
23. Almeida-Filho N. Higher education and health care in Brazil. Lancet. 2011;377(9781):1898-1900. https://doi.org/10.1016/s0140-6736(11)60326-7
24. Aguiar CM, Pessoa MA, Câmara AC, Perrier RA et al. Factors involved in the choice of dentistry as an occupation by Pernambuco dental students in Brazil. J Dent Educ. 2009;73(12):1401-1407. https://pubmed.ncbi.nlm.nih.gov/20007496/
25. Orsini C, Binnie V, Evans P, Ladezma P et al. Psychometric validation of the Academic Motivation Scale in a dental student sample. J Dent Educ. 2015;79(8):971-981. https://pubmed.ncbi.nlm.nih.gov/26246537/

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Effect of different toothpastes on permeability and roughness of eroded dentin

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ABSTRACT

Dentin hypersensitivity (DH) is characterized by rapid, acute pain arising from exposed dental tubules. ***Aim:*** *the aim of this study was to evaluate the roughness, tubule occlusion, and permeability of eroded dentin brushed with different toothpastes.* ***Materials and Method:*** *ninety bovine teeth were cut into blocks. Thirty hemifaces were protected with varnish and the other sixty were submitted to permeability tests. Specimens were divided into groups according to the dentifrices: without fluoride (WF), sodium fluoride (NaF), and stannous fluoride (SnF_2). The blocks were subjected to a 5-day erosive-abrasive protocol. Surface roughness and dentinal tubule occlusion (n=10) were assessed for both control and test hemifaces of the same sample along with permeability analysis (n=20). Two-way RM ANOVA and Tukey's post-hoc test were performed ($p \leq 0.05$).* ***Results:*** *NaF and SnF_2 presented higher roughness than WF. The number of open tubules was higher in WF. Permeability was higher in SnF_2, but there was no significant difference between WF and NaF.* ***Conclusions:*** *both fluoride toothpastes occluded dentinal tubules and increased roughness. NaF toothpaste promoted greater decrease in dentin permeability.*

Keywords: *dentifrices - dentin desensitizing agents - dentin permeability - tooth abrasion - tooth erosion*

Efeito de diferentes cremes dentais na permeabilidade e rugosidade da dentina erodida

RESUMO

A hipersensibilidade dentinária (HD) é caracterizada por dor rápida e aguda decorrente de túbulos dentais expostos. ***Objetivo:*** *este estudo teve como objetivo avaliar a rugosidade, oclusão tubular e permeabilidade da dentina erodida escovada com diferentes dentifrícios.* ***Materiais e Método:*** *noventa dentes bovinos foram seccionados em blocos. Trinta hemifaces foram protegidas com verniz e outras sessenta foram submetidas à permeabilidade. Os espécimes foram divididos em grupos de acordo com os dentifrícios: sem flúor (SF), fluoreto de sódio (NaF) e fluoreto estanoso (SnF_2). Em seguida, os blocos foram submetidos a um protocolo erosivo-abrasivo de 5 dias. A rugosidade da superfície e a oclusão do túbulo dentinário (n = 10) foram avaliadas para ambas as hemifaces de controle e teste da mesma amostra, também realizou-se a análise de permeabilidade (n = 20). Two-way RM ANOVA e pós-teste de Tukey foram realizados ($p \leq 0,05$).* ***Resultados:*** *NaF e SnF_2 apresentaram rugosidade superior ao SF. O número de túbulos abertos foi maior em SF. Não encontramos diferenças significativas entre SF e NaF em relação à permeabilidade; entretanto, SnF_2 apresentou maior permeabilidade.* ***Conclusão:*** *ambos os dentifrícios fluoretados foram capazes de ocluir os túbulos dentinários e aumentar a rugosidade. No entanto, o dentifrício NaF promoveu uma maior diminuição da permeabilidade dentinária.*

Palavras-chave: *dentifrícios - agentes dessensibilizantes da dentina - permeabilidade da dentina - abrasão dentária - erosão dentária*

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INTRODUCTION

Dentin hypersensitivity (DH) is characterized by rapid, acute pain arising from exposed dental tubules in response to thermal, tactile, chemical, and osmotic stimuli[1]. According to the hydrodynamic theory, physical stimuli promote the movement of fluids within dentin tubules, contracting and expanding odontoblastic processes and stimulating nerve fibers present in the dentin-pulp interface[2].

DH affects patients' oral health and quality of life, and is one of their main complaints[1]. The etiology of DH is multifactorial and often associated with non-carious cervical lesions (NCCL)[1,3], whose prevalence is about 11.5%, regardless of the etiology. Molars and premolars are the teeth most frequently affected by NCCL, with incidence and severity increasing with patient age[4]. The depth and morphology of lesions contribute to high DH levels, and are affected by gastric disease and acidic beverage intake[3].

Dental erosion is one of main factors involved in DH. It results from the loss of surface tooth tissue due to the action of acids from intrinsic or extrinsic sources[5,6]. Acids from intrinsic sources affect the palatal surface of anterior teeth and the occlusal surface of posterior teeth, whereas those from extrinsic sources affect the vestibular and cervical surfaces of anterior teeth[6]. The action of acids and proteases changes to dissolve minerals, glycoproteins and other components responsible for occluding dentinal tubules[7]. Reduced salivary flow, either due to medication use or xerostomia, makes it difficult to restore the oral pH and, consequently, contributes to the erosive process[3].

Desensitizing and anti-erosive toothpastes are indicated for treating DH[8,9]. However, their beneficial effects are limited, with results ranging from no action to almost complete desensitization and erosion inhibition, depending on the type. High-quality studies are therefore needed[2].

Different components have been added to toothpastes with the aim of increasing the resistance of the dental substrate to erosive processes and/or dentin tubule obliteration[2,10,11]. Dentifrices containing polyvalent metal ions, such as stannous ions, have effectively reduced the erosion process by depositing ions on eroded tissue, thereby preventing contact with acids[2,10,12,13,14]. Conversely, abrasive compounds are also often added to dentifrices to improve their cleaning potential[15], which may influence the exposure of dentin tubules after erosion-abrasion cycles, increasing permeability and roughness, which have been associated with the development of DH[16,14]. Although some studies have evaluated dentifrice action on eroded dentin[17,18], there are few studies that have simultaneously evaluated roughness, permeability, and tubular obliteration with the use of sodium fluoride- and stannous fluoride-based dentifrices. The aim of this study was therefore to evaluate eroded dentin properties after erosive-abrasive challenge using three different commercial dentifrices. The hypothesis was that there are no differences between dentifrices in protecting against erosive-abrasive cycles regarding roughness, tubular obliteration, and dentin permeability.

MATERIALS AND METHODS

The project was approved by the Ethics Committee on the Use of Animals of Araçatuba School of Dentistry (#00414-2018).

Experimental design

The following factors were assessed: (1) two kinds of bovine dentin (sound and eroded); and (2) three dentifrices – fluoride free (without fluoride - WF-negative control – Curaprox Enzycal Zero rybol AG Neuhausen as Rheinfall. Switzerland); with sodium fluoride (NaF - positive control, 1450 ppm NaF Colgate Total 12, Colgate Palmolive, São Bernardo do Campo, SP, Brazil); and with stannous fluoride (SnF_2, 1100 ppm F as SnF_2 Crest Pro-Health, P&G, Cincinnati, Ohio, USA). Table 1 shows the composition and characteristics of each dentifrice.

Response variables were surface roughness, dentinal tubule occlusion assessed using scanning electron microscopy (SEM), and dentin permeability. Figures 1 and 2 shows flowcharts of the experiments.

Preparation of dentin specimens

Ninety bovine incisors from 23 animals aged 24 to 30 months were extracted, cleaned, and stored in distilled water until the beginning of the experiment, for a maximum time of 60 days. Any teeth with clinical evidence of caries, root resorption, cracks, or fractures were excluded from the study. Dental crowns were separated from the roots using an IsoMet 1000 precision cutting saw (Buheles, IL, USA) with diamond disc (4" × 0.12" × ½", Buehler, Illinois, USA) under water cooling.

Table 1. Dentifrices used in the study and their composition according to manufacturers.

Material	Tipe	Code	Composition	Manufacture
Curaprox Enzycal Zero (RDA-60) † Batch:442MHDEXP1121	Fluoride-free Toothpaste	WF	Water, Sorbitol, Hydrated Silica, Glycerin, Steareth-20, Titanium Dioxide (Cl 77891), Aroma, Sodium Phosphate, Carrageenan, Sodium Chloride, Citric Acid, Sodium Benzoate, Potassium Thiocyanate, Glucose Oxidase, Amyloglucosidase, Lactoperoxidase	Trybol AG, Neuhausen AM Rheinfall, Swiss.
Colgate Total 12 (RDA-70/80) † Batch:6184BR121R	Sodium Fluoride Toothpaste	NaF	Sodium Fluoride (1450 ppm as NaF) Water, Triclosan, Sorbitol, Silica, Sodium Lauryl Sulfate, PMV / MA Copolymer, Sodium Hydroxide, Saccharin Sodium, Titanium Dioxide	Colgate-Palmolive, São Bernardo do Campo, SP, Brazil.
Crest Pro-Health (RDA-155) † Batch:6039GF	Stannous Fluoride Toothpaste	SnF2	Stannous fluoride (1100 ppm F as SnF2) Glycerin, Hydrated Silica, Sodium Hexametaphosphate, Propylene Glycol, PEG 6, Water, Zinc Lactate, Trisodium Phosphate, Sodium Lauryl Sulfate, Sodium Lauryl Sulfate, Carrageenan, Sodium Saccharin, Xanthan Gum, Blue 1	P&G, Cincinnati, Ohio, USA.

†RDA values according to manufacturers

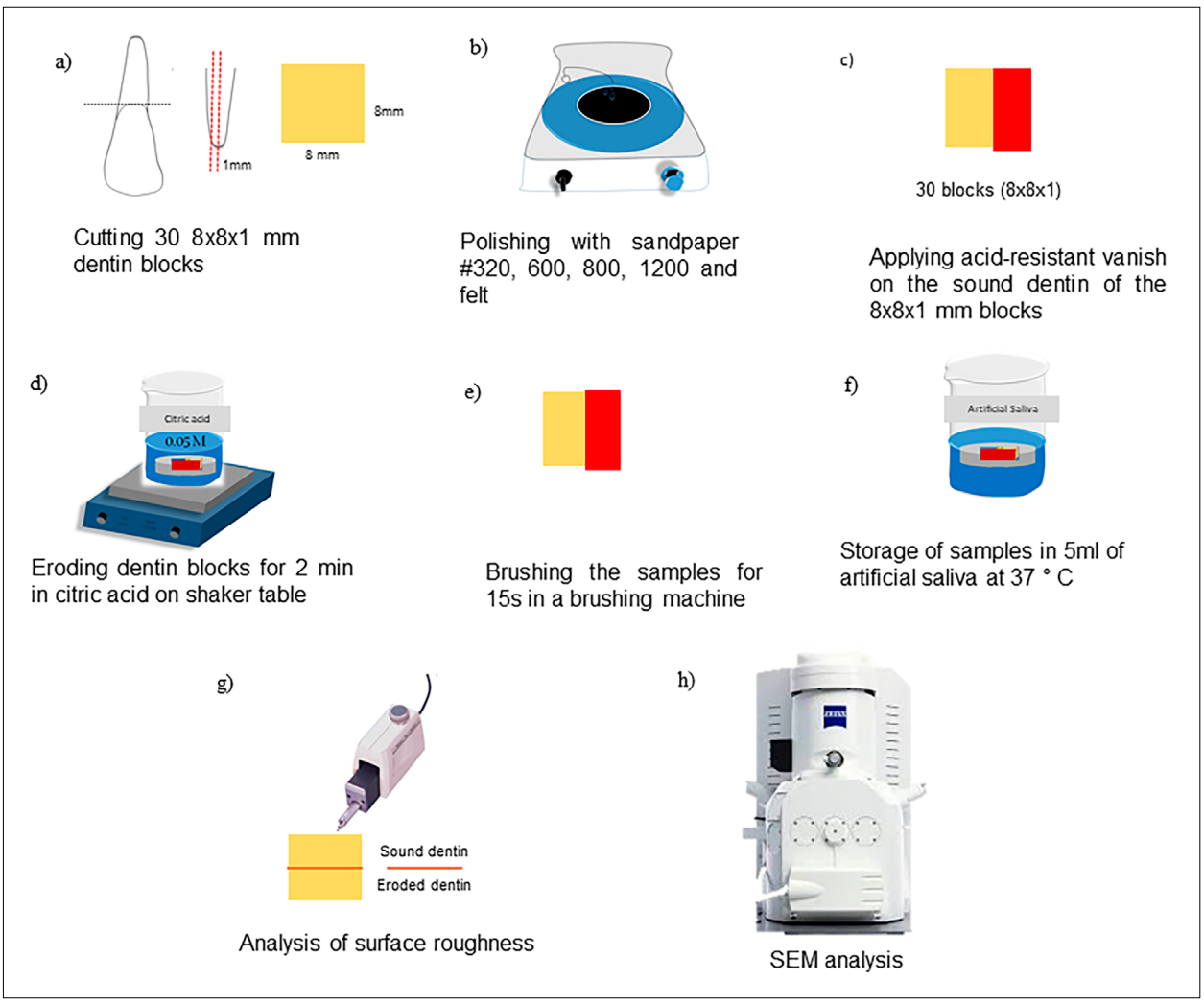

Fig. 1: *Flowchart showing the steps followed to analyze roughness and tubule obliteration*

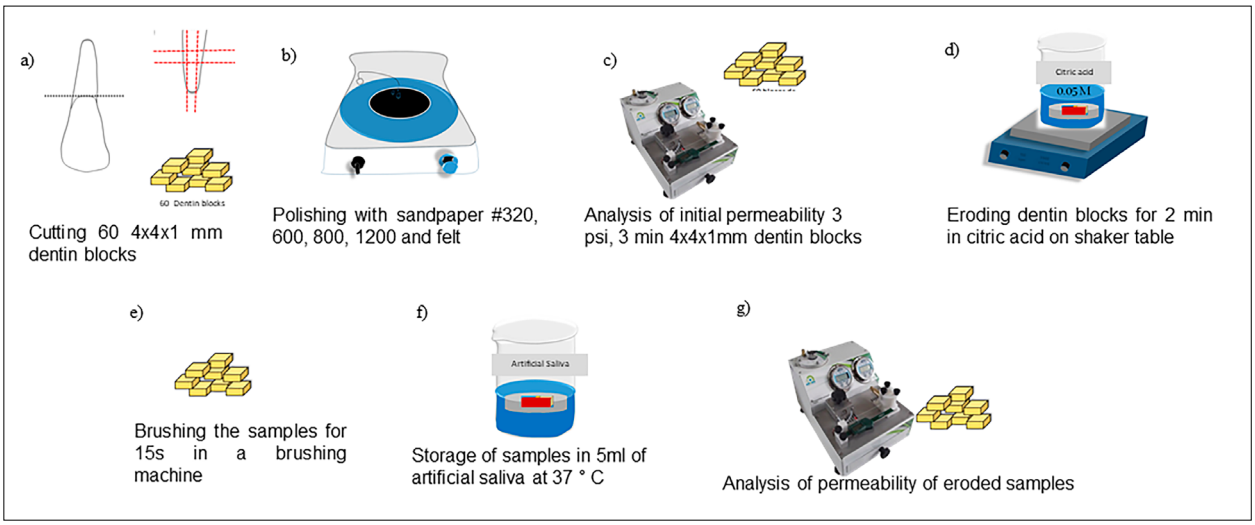

Fig. 2: *Flowchart showing the steps followed to analyze dentin permeability*

Sixty roots were cut with diamond disc (4" × 0.12" × ½", Buehler, Illinois, USA) under water cooling to obtain dentin blocks (4 × 4 × 1 mm), and abraded with sandpaper discs of decreasing grit (# 320, # 600, # 800, and #1200) until they were 1 mm thick, and prepared to undergo initial dentin permeability analysis.

The other thirty roots were sectioned transversely into blocks 1 mm thick (8 × 8 mm), which were polished with silicon carbide sandpaper discs of decreasing grit (# 320, # 600, # 800, and # 1200) using a polisher (Aropol E, Arotec, Cotia, SP, Brazil). The final polishing was performed using felt disc and diamond polishing paste (1 µm, Arotec APL4), and surfaces were coated with an acid-resistant varnish (Colorama, São Paulo, SP, Brazil), to serve as a reference area (sound dentin).

Initial analysis of dentin permeability

To standardize dentin blocks, the 60 specimens (4 × 4 × 1mm) underwent initial dentin permeability analysis. Specimens were immersed in a 37% phosphoric acid solution (1 ml/block) for 30 seconds, washed twice for 1 minute in deionized water, and dried with absorbent paper to remove the smear layer. Cellular and extracellular contents were removed from inside dentinal tubules by immersing the blocks in a 10% NaOH solution (3 ml/block) for 6 hours, followed by deionized water (3 ml/block) for a further 6 hours[19,20].

Each specimen was coupled to a fluid infiltration system (Dentin Permeability, Odeme, Luzerna, SC, Brazil) with 1.9 psi initial water pressure. An air bubble was inserted in the capillary tube, and the pressure was increased to 3 psi, stabilizing the air bubble for 30 seconds. After stabilization, the flow through the block was recorded for 3 minutes by recording the bubble displacement within the capillary tube using a 1 mm-resolution digital caliper. The displacement, expressed in mm, provides the flow rate (Q) – volume of deionized water passing through dentinal tubules –, determined by the following formula: $Q = (Vs \times D)/(L \times T)$, where Vs is the standard volume of the capillary tube in µl, D the bubble displacement in mm, L the capillary length in mm, and T the time in minutes. The dentin permeability (Lp), expressed in $\mu l.cm^2/min.cm\ H_2O$, is calculated based on the filtration rate (Q), using the following formula: $Lp = Q/(PH \times Asup)$, where PH is the hydrostatic pressure and Asup the exposed dentin surface area[16,20]. Specimens with initial dentin permeability between 1.5 and 2.5 $\mu l.cm^2/min.cm\ H_2O$ were selected.

Erosion – abrasion process

For the erosion process, thirty blocks (8 × 8 × 1 mm) were coated with an acid-resistant varnish (Colorama, São Paulo, SP, Brazil), serving as reference area (sound dentin) prior to the challenge. The other sixty blocks (4 × 4 × 1 mm) underwent the challenge after initial permeability had been determined. The erosion process consisted of treating the specimens using an orbital shaker table (Tecnal TE – 420, Piracicaba, SP, Brazil), while immersed in 0.05 M citric acid solution (Merk Biochemistry, Damstadt, Germany) (pH 3.2 – 2 ml/specimen) for 2 minutes, 4 times per

day, with 1-hour intervals between each cycle, for 5 days[21]. After the first and last erosive cycles of the day, specimens were brushed with the evaluated toothpastes for 15 seconds (1:3 – toothpaste/distilled water – slurry) using an automatic brushing machine, 45 strokes, 2 N force (MSET, Elquip, São Carlos, SP, Brazil) perpendicular to the specimen surface (extra-soft Condor, round bristles, São Bento do Sul, SC, Brazil) and immersed in slurry for 2 minutes at room temperature[21]. After each cycle, specimens were rinsed in distilled water for 30 seconds and stored in a remineralizing solution (artificial saliva: 1.5 mmol.l^{-1} $Ca(NO_3)_2 \cdot 4H_20$; 0.9 mml.l^{-1} $NaH_2PO_4.2H_2O$; 150 mmol.l^{-1} KCl, 0.1 mol.l^{-1} Tris buffer; pH 7.0) at 37 °C. Specimens were kept in artificial saliva at 37 °C at the end of each experimental day and stored in 100% humidity, with distilled water at 37 °C at the end of the erosive-abrasive protocol until they were analyzed.

Analysis of surface roughness

Surface roughness was analyzed on the 8 × 8 × 1 blocks. Each specimen presented half sound and half eroded dentin - both analyzed after cycling (Surftest SJ 401 Rugosimeter - Mitutoyo, Mitutoyo American Corporation, Aurora, Illinois, USA) according to the Ra roughness pattern, which is the arithmetic mean between peaks and valleys. The cut-off used to maximize surface undulation filtering was 0.25 mm. Three equidistant readings were made on each surface and averaged[22].

Analysis of dentinal tubule obliteration

Three specimens (8 × 8 × 1 mm) per group were fixed in gold-covered metal stubs (SCD 050, Balzers) and subjected to scanning electron microscopy (EVO HD LS-15, Carl Zeiss do Brasil Ltda, SP, Brazil), at 15 kV and ×2000 magnification. The number of obliterated tubules was calculated for both sound and eroded surfaces in the same specimen, starting at the center of the sample and following northwest and southeast directions. A representative image of the specimen was used for quantitative analysis of dentinal tubules using the ImageJ software (National Institutes of Health, Betseda, MD, USA). Total number of tubules was determined by counting all the tubules present in the image (Fig. 3A), and the percentage of unblocked tubules was calculated[23-24].

Final analysis of dentin permeability

The final analysis of dentin permeability was performed after the erosive-abrasive cycles, as described above.

Statistical analysis

Surface roughness, tubular obliteration, and dentin permeability showed normal distribution

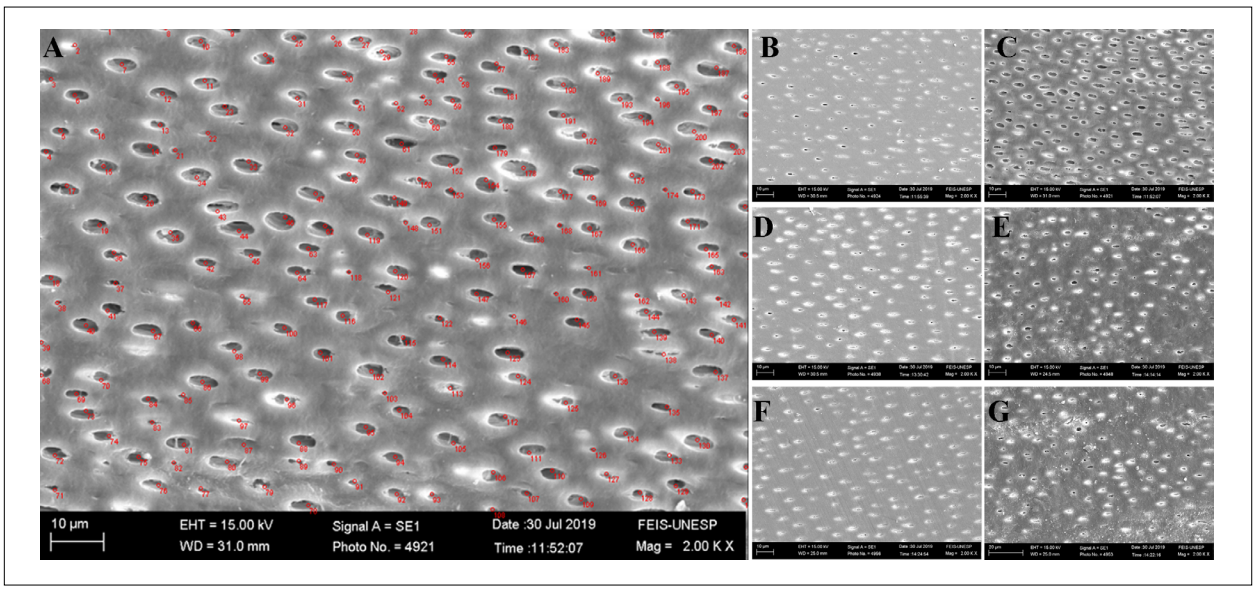

Fig. 3: *Example of Image J software for counting open tubules. Dentin surface on the sound and eroded dentin.A: Tubule count B: WF sound dentin group; C: WF eroded dentin group; D: NaF sound dentin group; E: NaF eroded dentin group; F: SnF_2 sound dentin group; G: SnF_2 eroded dentin group.*

(Shapiro-Wilk test) and homogeneity (Cochran test). Data on bovine dentin (sound and eroded) and dentifrices were submitted to two-way repeated measures ANOVA, followed by Tukey's post-hoc test. Statistical analysis was performed using the SigmaPlot 12.0 software (System Software, San Jose, CA, USA), with 5% significance level.

RESULTS

Figure 4 presents the results of the surface roughness analysis (Ra). The WF toothpaste showed the lowest roughness after the erosive-abrasive cycles (p=0.001), with values significantly different from those recorded for the fluoride toothpastes, which were statistically similar to each other (p=0.735). After the erosive-abrasive cycles, all the tested toothpastes promoted a significant increase in dentin surface roughness (p<0.001).

Figures 3 and 5 show the results of the analysis of dentin tubule obliteration using scanning electron microscopy (SEM). No difference was found between toothpastes for sound dentin (p≥0.05); however, the WF group had a significantly greater number of unblocked dentin tubules (84%) after the erosive-abrasive cycles compared to the NaF (49.2%) and SnF_2 (61.4%) groups (p<0.001), which were statistically similar (p=0.201). All groups presented a significantly increased number of unblocked tubules after the erosion-abrasion cycles (p≤0.05). Figure 3 shows an alteration in specimen surface induced by the action of citric acid. In the group comparison, WF presented the greatest number of open dentinal tubules and SnF_2 presented some granules deposited on the surface.

Figure 6 shows the results of dentin permeability.

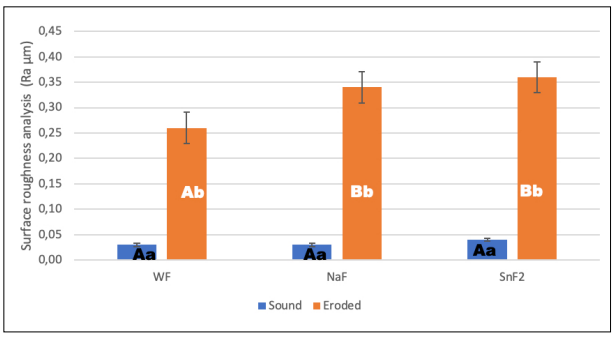

Fig. 4: *Surface roughness analysis (Ra µm) of dentin blocks according to the treatment group and the specimen area (sound and eroded). Different letters (uppercase between groups and lowercase intragroup) indicate statistically significant differences (p <0.05). WF: toothpaste without fluoride. NaF: toothpaste with sodium fluoride. SnF2: toothpaste with stannous fluoride.*

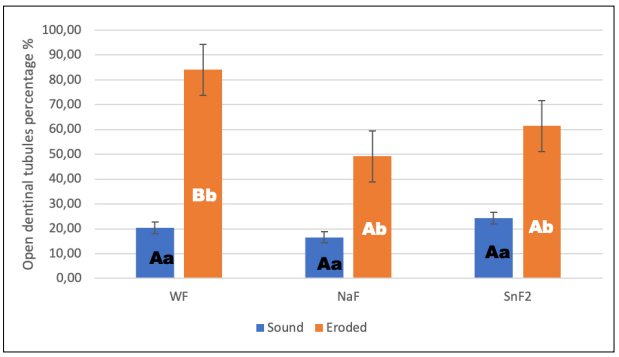

Fig. 5: *Dentinal tubules occlusion – SEM evaluation: values of open dentinal tubules, as a percentage of sound and eroded dentin. Different letters (uppercase between groups and lowercase intragroup) indicate statistically significant differences (p <0.05). WF: toothpaste without fluoride. NaF: toothpaste with sodium fluoride. SnF2: toothpaste with stannous fluoride.*

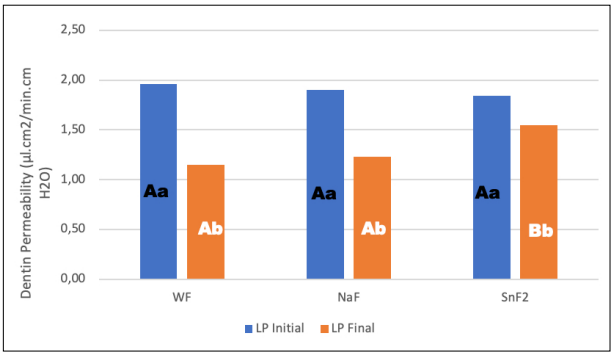

Fig. 6: *Dentin permeability according to dentifrice (µl.cm²/min.cm H2O). Different letters (uppercase between groups and lowercase intragroup) indicate statistically significant differences (p <0.05). Lp = dentin permeability. WF: toothpaste without fluoride. NaF: toothpaste with sodium fluoride. SnF2: toothpaste with stannous fluoride.*

No statistical difference was found between WF and NaF toothpastes (p=0.912, 45% and 40% reduction, respectively), but SnF_2 showed a 14% reduction in dentin permeability after erosive-abrasive cycles when compared to the other dentifrices (p<0.001). All three dentifrices decreased dentin permeability after the erosive-abrasive cycles (p<0.001).

DISCUSSION

Since one of the main etiologic factors of dentin hypersensitivity (DH) is erosion[5], various studies have analyzed several types of dentifrices with desensitizing and anti-erosive components to evaluate their action on the dentin surface[2,8,9,17,18,25-27]. In the current study, in order to ascertain whether dentifrice fluoride content affected the results, a toothpaste without fluoride (WF) was selected as a negative control. The toothpaste containing sodium

fluoride (NaF) was selected as a positive control based on studies that report its protective capacity on the eroded tooth surface[10,18,25,28]. The toothpaste containing stannous fluoride (SnF_2) was selected because studies indicate that its components may cause superficial obliteration of dentinal tubules[2,18,27,29], thus addressing the need to assess tubular obliteration and dentin permeability[11,16,24]. Surface roughness was analyzed to evaluate the influence of toothpaste abrasiveness on the surface of the eroded and abraded substrates.

Dentifrices contain abrasive particles, mostly silica derivatives, as polishing agents[15,30]. These materials are added to toothpastes to eliminate bacterial biofilms and reduce other debris accumulation on tooth surfaces, but they may increase dentin wear by increasing toothpaste slurry abrasivity (relative dentin abrasivity or RDA)[30,31]. The WF toothpaste caused significantly lower roughness than others, probably due to its lower RDA value[32]. The NaF and SnF_2 dentifrices promoted similar roughness and wear on eroded dentin, which may be explained by the amount of silica and the chemical influence of other components present in the NaF-based toothpaste[10,30]. After simulating toothbrushing abrasion equivalent to two years, Aguiar et al.[32] found that dentifrices with RDA values similar to those of SnF_2-based toothpastes cause roughness similar to that caused by the NaF dentifrice, in agreement with our results[32].

Surface roughness was higher in eroded dentin than in sound dentin for all tested toothpastes. Tooth surfaces with values above 0.2 and 0.3 µm (after cycling) increased bacterial adhesion to dentin surface[33]. The literature reports that toothpastes may increase surface roughness 8-fold[32,33]. This finding corroborates our results, where roughness increased by 8 to 10 times as specimens underwent both abrasive and erosive action.

In addition to silica, toothpastes with desensitizing or anti-erosive action contain particles capable of obliterating dentinal tubules, which can be achieved either by an occluding layer on top of the dentin or by depositing material inside the tubules[25]. Scanning electron microscopy (SEM) showed a high percentage of open dentinal tubules after the erosive-abrasive challenge, especially in the WF group, mainly because toothpaste WF does not contain functional agents capable of inducing tubular occlusion or intratubular mineralization[25]. Studies on SnF_2-based toothpastes have reached controversial results, since some found it to be superior to conventional toothpastes[12,13,14,29], whereas others reported similar results between them[25,28], as does our study. Some studies reported the presence of SnF_2 particles on the substrate surface, obliterating the top of dentinal tubules[18,25], but found no particles within tubules when they were analyzed in cross-sections[25]. According to a study by João-Souza et al.[34], stannous has no protective effect against dental erosion and abrasion but helps reduce open tubular areas.

The current study also verified a significant increase in the number of open tubules for both sound and eroded dentin in all groups. This is due to exposure to citric acid. Frequent consumption of citric acid leads to tooth structure loss and smear layer removal[16]. Using an experimental dentifrice with sodium trimetaphosphate associated with fluoride, Favretto et al.[19] found a greater number and larger internal area of open tubules by SEM analysis in placebo groups subjected to erosive challenges, as occurred in the WF group in our study[19].

Although SnF_2 caused dentin obliteration similar to that of NaF, it promoted a significantly higher permeability of eroded dentin compared to the other dentifrices after cycling. This may be due to tissue wear resulting from toothpaste abrasiveness, leading to a loss of substrate thickness and possibly interfering with its desensitizing effect[25]. According to Arnold et al.[25], a dentin thickness of approximately 61.98 µm may be lost after an erosive cycle performed with highly abrasive stannous-based dentifrices, which may contribute to dentin sub-surface analysis. Another study found that a SnF_2 dentifrice was unable to reduce dentin permeability, in contrast to the NaF dentifrice and two other toothpastes with desensitizing action (one containing calcium silicate and sodium phosphate, and another with potassium nitrate)[17,25]. Dentinal obliteration can be dissolved by acids[25], and the high concentration of abrasive components in SnF_2 dentifrices may favor stannous ion bonding with silica, reducing its anti-erosive action[15]. A meta-analysis found that SnF_2-based toothpaste achieved better outcomes than other compounds within a period of up to 2 weeks. However, it lost effectiveness at the 4 and 8-week follow-up, presenting a rapid desensitizing effect[8]. Another systematic review found that toothpastes containing SnF_2 reduced DH effects after 8 to 15 days of brushing[27]. The use of artificial saliva and

the absence of dental biofilm and salivary film may reduce fluoride retention on surfaces in 5-day *in vitro* erosive protocols[35], which are supposedly more aggressive than an *in vivo* cycling protocol for the same period.

Although fluoride has beneficial effects in treating caries lesions, its actions on the erosive process are not as noticeable[26]. In the current study, the NaF-based dentifrice contains 350 ppm more fluoride (F) than SnF_2. Despite the reduced action of fluoride in the erosive process, this difference may have influenced the lower permeability found in the SnF_2 group in relation to NaF. These results contradict those reported in another study that evaluated NaF and SnF_2 dentifrices, which found decreased dentin permeability for both[18].

The group treated with WF toothpaste showed a greater number of open tubules, but lower permeability in eroded dentin, which may be explained by the deposition of calcium phosphate from saliva and a smear layer on the dentin surface[20,36]. From the results of a meta-analysis that evaluated 30 randomized clinical trials, it was concluded that conventional fluoride toothpastes showed no significant difference in desensitizing effect when compared to placebo[8]. According to João-Souza et al.[34], the abrasion resulting from toothbrushing on demineralized dentin may play a role in tubal occlusion by forming a smear layer on the dentin surface. This accounts for the significantly reduced permeability after the erosive-abrasive challenge for all groups when comparing sound and eroded dentin. According to the literature, reduction in dentin permeability ranges from 5 to 50% after treatment with different anti-erosive dentifrices[17,36]. In the current study, permeability decreased by 14 to 45%, considering all evaluated toothpastes.

This study has some limitations, such as the fact that it did not identify the composition of the particles in the top of the dentinal tubules. The toothpastes evaluated contain other components besides fluoride compounds, which may influence the results. The current study chose to evaluate commercially available toothpastes in order to represent real-life conditions. Further *in situ* and *in vivo* studies evaluating the effectiveness of different toothpastes are needed to verify the influence of human saliva on eroded dentin, seeking to prolong the effects of toothpastes on dentinal tubule occlusion for the treatment of DH.

CONCLUSIONS

Our findings show that the two fluoride-based dentifrices are equally able to occlude dentinal tubules and increase the roughness of the eroded dentin, and that the sodium fluoride toothpaste promoted a greater decrease in dentin permeability.

CONFLICT OF INTEREST DISCLOSURE

The authors declare no potential conflicts of interest regarding the research, authorship, and/or publication of this article.

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REFERENCES

1. Goh V, Corbet EF, Leung WK. Impact of dentine hypersensitivity on oral health-related quality of life in individuals receiving supportive periodontal care. J Clin Periodontol 2016;43:595-602. https://doi.org/10.1111/jcpe.12552
2. Hu ML, Zheng G, Zhang YD, Yan X, et al. Effect of desensitizing toothpastes on dentine hypersensitivity: a systematic review and meta-analysis. J Dent 2018;75:12-21. https://doi.org/10.1016/j.jdent.2018.05.012
3. Teixeira DNR, Zeola LF, Machado AC, Gomes RR, et al. Relationship between non carious cervical lesions, cervical dentin hypersensitivity, gingival recession, and associated risk factors: a cross-sectional study. J Dent 2018;76:93-97. https://doi.org/10.1016/j.jdent.2018.06.017
4. Zeola FL, Soares PV, Cunha-Cruz J. Prevalence of dentin hypersensitivity: systematic review and meta-analysis. J Dent 2019;81:1-6. https://doi.org/10.1016/j.jdent.2018.12.015
5. Ganss C. Is erosive tooth wear and oral disease? Monogr Oral Sci 2014;25:16-21. https://doi.org/10.1159/000359931
6. Ganss C, Lussi A, Schlueter N. The histological features and physical properties or eroded dental hard tissues. Monogr Oral Sci 2014;25:99-107. https://doi.org/10.1159/000359939
7. Kanzow P, Wegehaupt FJ, Attin T, Wiegand A. Etiology and pathogenesis of dental erosion. Quintessence Int. 2016;47:275-278. https://doi.org/10.3290/j.qi.a35625
8. Hu ML, Zheng G, Lin H, Yang M, et al. Network meta-analysis on the effect of desensitizing toothpastes on dentin hypersensitivity. J Dent 2019;88:103170. https://doi.org/10.1016/j.jdent.2019.07.008
9. Moraschini V, da Costa LS, dos Santos GO. Effectiveness for dentin hypersensitivity treatment of non-carious cervical lesions: a meta-analysis. Clin Oral Investig 2018;22:617-631. https://doi.org/10.1007/s00784-017-2330-9

10. Aykut-Yetkiner A, Attin T, Wiegand A. Prevention of dentine erosion by brushing with anti-erosive toothpastes. J Dent. 2014;42:856-861. https://doi.org/10.1016/j.jdent.2019.07.014
11. Pinto SC, Bandéca MC, Pinheiro MC, Cavassim R, et al. Preventive effect of a high fluoride toothpaste and arginine-carbonate toothpaste on dentinal tubules exposure followed by acid challenge: a dentine permeability evaluation. BMC Res Notes 2014;7:385. https://doi.org/10.1186/1756-0500-7-385
12. Hines D, Xu S, Stranick M, Lavender S, et al. Effect of a stannous fluoride toothpaste on dentinal hypersensitivity: In vitro and clinical evaluation. J Am Dent Assoc 2019;150:S47-S59. https://doi.org/10.1016/j.adaj.2019.01.006
13. Mahmoodi B, Goggin P, Fowler C, Cook RB. Quantitative assessment of dentine mineralization and tubule occlusion by NovaMin and stannous fluoride using serial block face scanning electron microscopy. J Biomed Mater Res B Appl Biomater 2021;109:717-722. https://doi.org/10.1002/jbm.b.34737
14. Olley RC, Mohsen BM, Alhaij S, Appleton PL. Measurement of eroded dentine tubule patency and roughness following novel dab-on or brushing abrasion. J Dent 2020;98:103358. https://doi.org/10.1016/j.jdent.2020.103358
15. Ganss C, Möllers M, Schlueter N. Do abrasives play a role in toothpaste efficacy against erosion/abrasion? Caries Res 2017;51:52-57. https://doi.org/10.1159/000452867
16. Pinto SC, Batitucci RG, Pinheiro MC, Zandim DL, et al. Effect of an acid diet allied to sonic toothbrushing on root dentin permeability: an in vitro study. Braz Dent J 2010;21:390-395. https://doi.org/10.1590/s0103-64402010000500002
17. João-Souza SH, Scaramucci T, Bühler-Borges A, Lussi A, et al. Influence of desensitizing and anti-erosive toothpastes on dentine permeability: an *in vitro* study. J Dent 2019;89:103176. https://doi.org/10.1016/j.jdent.2019.07.014
18. Lucchese A, Bertacci A, Zanna S, Chersoni S, et al. In vitro effects of fluoride-based and desensitizing toothpastes on dentin permeability. J Biol Regul Homeost Agents 2018;32:51-58.
19. Favretto CO, Delbem ACB, Moraes JCS, Camargo ER, et al. Dentin tubule obliteration using toothpastes containing sodium trimetaphosphate microparticles or nanoparticles. Clin Oral Investig 2018;22:3021-3029. https://doi.org/10.1007/s00784-018-2384-3
20. Favretto CO, Delbem ACB, Toledo PTA, Pedrini D. Hydraulic conductance of dentin after treatment with fluoride toothpaste containing sodium trimetaphosphate microparticles or nanoparticles. Clin Oral Investig 2021;25:2069-2076. https://doi.org/10.1007/s00784-020-03516-w
21. Pini NIP, Lima DA, Lovadino JR, Ganss C, et al. In vitro efficacy of experimental chitosan-containing solutions as anti-erosive agents in enamel. Caries Res 2016;50:337-345. https://doi.org/10.1159/000445758
22. Neres ÉY, Moda MD, Chiba EK, Briso ALF, et al. Microhardness and roughness of infiltrated white spot lesions submitted to different challenges. Oper Dent 2017;42:428-435. https://doi.org/10.2341/16-144-L
23. Cunha SR, Garofalo SA, Scaramuccu T, Zezell DM, et al. The association between Nd: YAG laser and desensitizing dentifrices for the treatment of dentin hypersensitivity. Lasers Med Sci 2017;32:873-880. https://doi.org/10.1007/s10103-017-2187-9
24. Garofalo SA, Sakae LO, Machado AC, Cunha SR, et al. In vitro effect of innovative desensitizing agents on dentin tubule occlusion and erosive wear. Oper Dent 2019;44:168-177. https://doi.org/10.2341/17-284-L
25. Arnold WH, Prange M, Naumova EA. Effectiveness of various toothpastes on dentine tubule occlusion. J Dent 2015;43:440-449. https://doi.org/10.1016/j.jdent.2015.01.014
26. Lussi A, Carvalho TS. The future of fluorides and other protective agents in erosion prevention. Caries Res 2015;49:18-29. https://doi.org/10.1159/000380886
27. Marto C M, Baptista Paula A, Nunes T, Pimenta M, et al. Evaluation of the efficacy of dentin hypersensitivity treatments- A systematic review and follow-up analysis. J Oral Rehabil 2019;46:952-990. https://doi.org/10.1111/joor.12842
28. Sharma N, Roy S, Kakar A, Greenspan DC, et al. A clinical study comparing oral formulations containing 7.5% calcium sodium phosphosilicate (NovaMin), 5% potassium nitrate, and 0.4% stannous fluoride for the management of dentin hypersensitivity. J Clin Dent 2010;21:88–92.
29. Assunção CM, Schlueter N, Rodrigues JA, Carvalho TS, et al. Do fluoride toothpastes have similar preventive effect in permanent and primary teeth against erosive tooth wear? Int J Paediatr Dent 2018;29:228-236. https://doi.org/10.1111/ipd.12449
30. Johannsen G, Tellefsen G, Johannsen A, Liljeborg A. The importance of measuring toothpaste abrasivity in both a quantitative and qualitative way. Acta Odontol Scand 2013;71:508-517. https://doi.org/10.3109/00016357.2012.696693
31. Vertuan M, Souza BM, Machado PF, Mosquim V, et al. The effect of commercial whitening toothpastes on erosive dentin wear in vitro. Arch Oral Biol 2020;109:104580. https://doi.org/10.1016/j.archoralbio.2019.104580
32. Aguiar JD, Medeiros IS, Souza-Junior MHSE, Loreto SC. Influence of extended use of desensitizing toothpaste on dentin bonding microhardness and roughness. Braz Dent J 2017;28:346-353. https://doi.org/10.1590/0103-6440201601292
33. Cury MS, Silva CB, Nogueira RD, Campos MGD, et al. Surface Roughness and bacterial adhesion on root dentin treated with diode laser and conventional desensitizing agents. Lasers Med Sci 2018;33:257-262. https://doi.org/10.1007/s10103-017-2356-x
34. João-Souza SH, Sakae LO, Lussi A, Aranha ACC, et al. Toothpaste factors related to dentine tubule occlusion and dentine protection against erosion and abrasion. Clin Oral Investig 2020;24:2051-2060. https://doi.org/10.1007/s00784-019-03069-7
35. Alghilan MA, Cook NB, Platt JA, Eckert GJ, et al. Susceptibility of restorations and adjacent enamel/dentine to erosion under different salivary flow conditions. J Dent 2015;43:1476-1482. https://doi.org/10.1016/j.jdent.2015.10.007
36. Wang Y, Sa S, Sauro H, Chen W, et al. Effect of desensitizing toothpastes on dentinal tubule occlusion: a dentine permeability measurement and SEM in vitro study. J Dent 2010;38:400-410. https://doi.org/10.1016/j.jdent.2010.01.007

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LV REUNIÓN CIENTÍFICA ANUAL SAIO
30 - 31 OCT / 01 NOV 22
Buenos Aires ARGENTINA

SAIO
SOCIEDAD ARGENTINA DE INVESTIGACION ODONTOLOGICA
División Argentina de la International Association for Dental Research

On October 30-31 and November 1, 2022, the LV Annual Scientific Meeting of the Argentine Society of Dental Research was held at the Faculty of Dentistry of the University of Buenos Aires.

Dr. Pablo Rodriguez, Vice-President in charge of the Presidency of the Society, highlighted during his speech at the opening ceremony the fundamental role of the different research groups of the Society that develop tasks in diverse Universities all over the country. He also emphasized the need to promote the link between all members, highlighting a federal vision in the operation of the Institution and ensuring the allocation of resources to improve computer systems as a strategy to promote this communication. On the other hand, he stressed the importance of accompanying and supporting AOL, SAIO's official publication organ, in its growth.

The LV Annual Meeting had 334 registrants and received 178 abstracts and three research projects for presentation.

As part of the LV Annual Meeting, the following lectures were given:

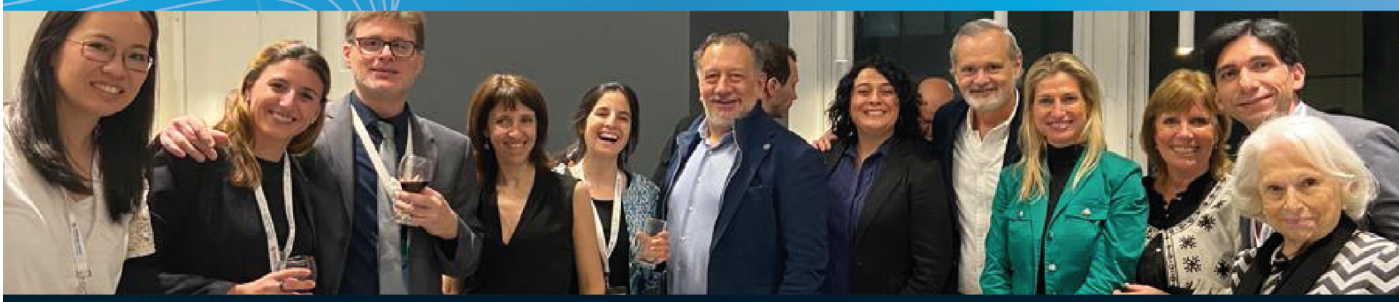
- "Dispenser for applying small standardized amounts of fluoride toothpaste in infants", by Dr. Silvia Chedid.
- "Strategies for research management in the dental area", by Dr. Pablo Alejandro Rodríguez.
- "Pillars of odontogeriatrics: proposal of a new philosophy", by Dr. Rodrigo A. Giacaman Sarah and Dr. Soraya León Araya.
- "Dentin hypersensitivity: evidence applied to epidemiology, aetiology and clinical management", dictated by Dr. Cassiano Kuchenbecker Rösing.

The symposium "Medical Cannabis in Dentistry" was also held, with the participation of Drs. Andrea De Laurentis, César Ángel Ossola and Mariana Ríos; as well as "Care and hygiene of removable dental appliances", coordinated by Dr. Luciana D' Eramo and Dr. Carla Masoli.

On the first day, the meetings of the Society's research groups were held: Pulp Biology and Regeneration, Cariology and Public Health, Education, Dental Materials, Oral Medicine and Pathology, Periodontics and Implants, Pediatric Dentistry and Orthodontics. The traditional presentation was also made by the Editorial Board of the Acta Odontológica Latinoamericana to communicate the current status of the journal.

A photography contest was organized to stimulate the dissemination of findings related to dental research in basic and clinical areas, and the following prizes were awarded:

- Divisional Award
  IADR Unilever Hatton Unilever Hatton Divisional Award
- Rodolfo Erausquin Award
- María Inés Egozcue Award
- Colgate Palmolive Award (Master and PhD students)
- GSK Award
- Omar Tumilasci Award
- María Luisa Rins de David Award
- Suzel M. Scozzarro Award
- FOUBA 75th Anniversary Award
- National Academy of Dentistry Award
- Aníbal Cobanera Oral Health Award
- Federa Award (Periodontics and Implants group)
- Dental Education Award (Education group)
- Prof. Dr. Héctor R. Maddalena Award (Dental Materials group)
- Orthodontics Award (Orthodontics group)



Members of the Board of Directors of SAIO and of the Organizing Committee together with the President of the Society, Dr. Pablo Rodríguez and the President of the Organizing Committee, Dr. Javier Fernández Solari.

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DIVISIONAL-IADR UNILEVER HATTON DIVISIONAL AWARD

"Glucocorticoid-induced osteoporosis: Osteocyte-mediated role in alveolar bone and tibia. "

Authors: Juliana Rodríguez*, Déborah Ruth Tasat, Carola Bettina Bozal .

DIVISIONAL-IADR UNILEVER HATTON DIVISIONAL AWARD

"Colonization of opportunistic pathogens in the buccal mucosa of children from vulnerable neighborhoods in Buenos Aires."

Authors: Andrea Muiño*, Isabel Adler, Silvia Aguas, Laura Harada, Mariana Díaz, Pablo Turón, Federico Galli, Pablo Rodríguez, Cristóbal Fresno, Valeria Denninghoff.

RODOLFO ERAUSQUIN AWARD

"Validation of the Cameriere method for estimating age of majority for forensic purposes. "

Authors: Ailin Gómez*, Stephanie Arario, Florencia Arrigo, María Julia Carosi, Carla Arias, Johanna Salazar, Pablo Salgado, Diego Vázquez, Luis Rannelucci, Briem Stamm Alan.

DR. MARÍA INÉS EGOZCUE AWARD

"Effect of silver diaminofluoride in a model of enamel demineralization. "

Authors: Yamila Sisca Jara*, Mariana Picca, Lidia Rocha Valadas, Pablo Salgado, Aldo Squassi, Luciana D´Eramo.

COLGATE-PALMOLIVE AWARD

"Study of the oral microbiota of infants younger than 24 months. "

Authors: Celina Cornejo*, Natalia Pin Viso, Susana Molgatini, Aldo Squassi, Laura Gliosca .

GSK AWARD

"Prevalence of taurodontia, dilaceration and angulation analysis of the upper 1st molar. "

Authors: Maria Julia Carosi*, Ailin Gomez, Alejandra Antoniuk, Florencia Arrigo, Stephanie Arario, Ariel Gualtieri, Sebastián Robledo, Diego Vázquez, Pablo Rodríguez .

OMAR TUMILASCI AWARD

"Application of cannabis oil (CBD:THC/1:1) to treat periodontitis. Preliminary study. "

Authors: Julieta Rodas*, Noelia Balcarcel, Gastón Troncoso, Julia Astrauskas, Claudia Mohn, Juan Carlos Elverdin, César Ossola, Javier Fernández Solari.

MARÍA LUISA RINS DE DAVID AWARD

"Deleterious effect of chronic alcohol on bone and periodontal tissue. "

Authors: Gastón Troncoso*, Clarisa Bozzini, Ana Clara Casadoumecq, Federico Mucci, Juan Carlos Elverdin, Javier Fernández Solari, Claudia Mohn.

SUZEL M. SCOZZARO AWARD

"Proinflammatory mediators in placental explants exposed to crevicular to crevicular fluid of pregnant women."

Authors: Luciana Doga*,Brenda Lara, Ifiaqui Loureiro, Luciana DÉramo, Sofía Novoa, Guillermina Calo, Laura Gliosca, Pablo Fabiano, Vanesa Hauk, Aldo Squassi, Claudia Perez Leiros.

75 YEARS FOUBA AWARD

"Immunohistochemical evaluation of odontogenic ghost cell lesions."

Authors: Lucas Polti*, María Luisa Paparella.

NATIONAL ACADEMY OF DENTISTRY AWARD

"PR SCORE applied in apical microsurgery with bovine and porcine biomaterials. "

Authors: Pablo Alejandro Rodríguez*, María Lorena Cabirta, Eugenia Miklaszewski, Nicolás Aíñe, Germán Tirao, Cristobal Fresno, Isabel Adler, Valeria Denninghoff.

PROF. DR. ANÍBAL COBANERA AWARD

"Morbidity due to dental caries in infants 2 and 3 years of age. "

Authors: Gabriela Alvaredo*, Celina Cornejo, Ximena Pazos, Pablo Salgado, Graciela Klemonskis, Aldo Squassi.

FEDERA AWARD

"Laser application in patients with post-surgical nerve post-surgical nerve injuries. Post-treatment evaluation."

Authors: Jesús Eduardo Fernández Alemán*, Ana Clara Casadoumecq, Sebastián Ariel Pura, Romina Chaintiou Piorno, María Agustina Seizar, David Heredia-Veloz, Ebingen Villavicencio-Caparo, Pablo Alejandro Rodríguez.

PROF. DR. HÉCTOR R. MADDALENA AWARD

"Microhardness of carious dentin treated with a 20% AGNCLS/PMAA solution. "

Authors: María Belén Cabalén*, Ignacio Mazzola, Laura Brain Lascano, Martin Sainz Aja, Mariana Picca, Gustavo Molina .

ORTHODONTICS AWARD

"Arch development with passive versus interactive self-ligating orthodontics. Clinical study."

Authors: Mahmood Ahmadi*, Andrea De Laurentis, Diana Calabrese, Marina Iglesias, Sandra Benitez Rogé, Alejandra Folco.