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# Evaluation of filling material removal in curved canals after different protocols: an ex vivo study with micro-computed tomography

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## ABSTRACT

Complete removal of the filling material is essential to the success of endodontic retreatment. However, this step can be challenging due to anatomical difficulties, the nature of the sealer, or operational factors.

**Aim:** To evaluate the efficacy of filling removal using ProDesign R and Reciproc Blue retreatment, with supplementary irrigation step with XP-Endo Finisher R. **Materials and Method:** Twenty mandibular molar mesial canals were prepared using ProTaper Next, and filled using the single-cone technique ( $n=10$ ). The fillings were subsequently removed using either ProDesign R (25/0.06) or Reciproc Blue (25/0.08 in the 3 mm from the tip of the file, decreasing towards the cervical, with average taper varying from 0.06 to 0.05 along the instrument). The XP-Endo Finisher R system was used for supplementary cleaning for both systems. Micro-computed tomography was used to assess filling material volume and reduction percentage. The significance level was set at 5% ( $p < 0.05$ ). **Results:** It was found that ProDesign R and Reciproc Blue removed 91.2% and 82.7% of the filling material, respectively ( $p > 0.05$ ), and after use of XP-Endo Finisher R, there was a significant increase in filling removal by 42.7 and 27.7%, respectively. **Conclusion:** ProDesign R and Reciproc Blue were equally effective, but neither system completely removed the filling material from the mesial canals of mandibular molars.

**Keywords:** endodontics - mandibular molar - micro CT - root canal therapy.

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## Avaliação da remoção de material de obturação em canais curvos após diferentes protocolos: um estudo ex vivo com microtomografia computadorizada.

## RESUMO

A remoção completa do material de obturação é essencial para o sucesso do retratamento endodôntico. Contudo, esta etapa pode ser desafiadora devido a dificuldades anatômicas, à natureza do cimento ou a fatores operacionais. **Objetivo:** Avaliar a eficácia da remoção de materiais de obturação utilizando os sistemas ProDesign R e Reciproc Blue para retratamento, com limpeza adicional empregando o sistema XP-Endo Finisher R. **Material e métodos:** Vinte canais mesiais de molares mandibulares foram preparados usando o sistema ProTaper Next e obturados pela técnica de cone único ( $n=10$ ). Os materiais de obturação foram subsequentemente removidos utilizando ProDesign R (25/0.06) ou Reciproc Blue (25/0.08 nos 3 mm da ponta da lima, diminuindo em direção ao terço cervical, com conicidade média variando de 0.06 para 0.05 ao longo do instrumento). O sistema XP-Endo Finisher R foi utilizado para limpeza suplementar em ambos os grupos. A microtomografia computadorizada (Micro-CT) foi empregada para avaliar o volume do material de obturação remanescente e sua porcentagem de redução. O nível de significância estatística foi estabelecido em 5% ( $p < 0.05$ ). **Resultados:** Constatou-se que ProDesign R e Reciproc Blue removeram 91,2% e 82,7% do material de obturação, respectivamente ( $p > 0.05$ ). Após o uso do XP-Endo Finisher R, houve um aumento significativo na remoção do material de obturação em 42,7% e 27,7%, respectivamente. **Conclusão:** ProDesign R e Reciproc Blue apresentaram eficácia similar, mas nenhum dos sistemas removeu completamente o material de obturação dos canais mesiais de molares mandibulares.

**Palavras-chave:** endodontia - molar mandibular - micro CT - tratamento do canal radicular.

## INTRODUCTION

Complete removal of filling material is essential to the success of endodontic retreatment<sup>1</sup>. Reciprocating instruments are used in endodontic retreatment because of their safety in extremely curved canals and their efficiency in removing filling material compared to continuous rotation systems or hand files<sup>2,3</sup>. A systematic review analyzed the effectiveness, efficiency, and apical extrusion of two continuous rotation systems (Protaper Universal and Mtwo retreatment systems) and two single-file reciprocating systems (Wave One and Reciproc) to determine which was the most efficient and effective and which extruded less filling material during endodontic retreatment. The authors concluded that all systems were equally time-efficient, but none completely removed the filling materials from straight root canals. In terms of apical extrusion, more material extruded towards the periapical tissues with the reciprocating systems than with the continuous rotation tested systems<sup>4</sup>.

Various studies have shown that complete removal of root canal filling material is not achieved<sup>5-7</sup>. Additional methods have been suggested to improve the removal of the remaining filling material, including passive ultrasonic irrigation and XP-endo finisher instruments<sup>8-11</sup>.

Many studies have demonstrated that micro-computed tomography (micro-CT) can be used to quantify filling material remaining in the root canal system before and after endodontic retreatment<sup>12-14</sup>. This technology provides accurate qualitative and quantitative three-dimensional analysis of root canal and filling material volume<sup>5</sup>.

Reciproc Blue (VDW GmbH, Munich, Germany) is a reciprocating single file system with a similar design to Reciproc, with an S-shaped cross-section and two cutting edges<sup>15,16</sup>. These files are available in sizes with tip diameters ranging from R25 to R50, all with a taper of 0.08 in the first 3 mm from the tip of the file, decreasing towards the cervical, with the average taper varying from 0.06 to 0.05 along the instrument. The technical features of Reciproc Blue provide superior performance in reciprocating movements, ensuring precision and safety during endodontic procedures<sup>17</sup>. It is made of blue thermomechanical-treated alloy, making it more flexible and resistant to flexural fracture<sup>15</sup>. Although initially developed for primary root canal treatment, its use for retreatment has been suggested<sup>18</sup>.

ProDesign R (Bassi/Easy, Belo Horizonte, Brazil) is an instrument designed for use in reciprocating motion. Featuring a double helix cross-section and CM thermal treatment for enhanced flexibility, it is available in lengths of 21 mm and 25 mm. Operating at 400 RPM with a 270° angle to the left and 30° to the right, its nickel-titanium alloy ensures precision and safety in dental procedures.

XP-Endo Finisher R (FKG Dentaire) is a 30-mm size non-tapered instrument, made of a NiTi MaxWire alloy. When inserted in the canal and exposed to body temperature, a martensitic-austenitic transformation occurs, and the instrument assumes a 'spoon shape' in the 10 mm segment from its tip. The instrument's expansion has the potential to reach anatomical areas that are inaccessible to conventional rotary instrumentation<sup>19</sup>. Recent studies have shown that the supplementary use of XP-Endo Finisher R (FKG) significantly reduced the amount of filling material in oval and curved canals re-treated with different systems<sup>19,20</sup>.

The aim of this study was to evaluate the efficacy of ProDesign R (PDR) and Reciproc Blue (RECB), followed by XP-Endo Finisher R, for filling material removal. The null hypothesis was that there would be no significant difference between the two systems in filling removal efficacy, and no significant difference in the amount of filling material remaining using the XP-Endo Finisher R.

## MATERIALS AND METHOD

G\*Power 3.1 software (Heinrich Heine College, Duesseldorf, Germany) was used to calculate power with a power of  $\beta = 95\%$  and  $\alpha = 5\%$ , and a t test for independent samples was applied. The ideal sample size for each group was found to be at least 10 teeth. Five additional samples per group were added to compensate for possible loss.

The study protocol was approved by the São Leopoldo Mandic Dental School Research Institute research ethics committee (CEP #2.332.649).

Ten human mandibular molars ( $n=10$ ) were obtained from a recently extracted tooth collection and stored in a 0.1% thymol solution. The inclusion criteria were teeth with two fully formed, independent mesial root canals, with separate foramina, and root curvature angles between 10° and 20°, confirmed by digital analysis of radiographs taken in the mesiodistal and buccolingual directions (Image J software,

Maryland, USA). Teeth with previous endodontic treatment, root resorptions, root fractures, or pulp calcifications were excluded. Specimen length was standardized to 18 mm using a diamond disk (FKG, La Chaux-de-Fonds, Switzerland). The canals were explored with a #10 K-type file (FKG, Chaux-de-Fonds, Switzerland), and the working length (WL) was set at 1 mm from the apical foramen.

### Root canal preparation

Initial root canal preparation was performed with the ProTaper Next until X2 file (25/0.06; Dentsply Sirona, Ballaigues, Switzerland) driven by a low-torque endodontic motor (X-Smart Plus; Dentsply Sirona) at a rotational speed of 300 rpm and a torque of 3 N.cm. Irrigation was performed with 5 mL of 2.5% sodium hypochlorite (NaOCl) followed by 5 mL of 17% ethylenediamine tetra-acetic acid (EDTA) for 3 minutes and then another 5 mL of 2.5% NaOCl.

The canals were dried with paper points (Dentsply Sirona). Each canal was then filled with gutta-percha and AH Plus sealer (Dentsply Sirona). Radiographs in the mesiodistal and buccolingual directions were used to confirm the final quality of the fillings. The teeth were sealed with the temporary filling material Coltosol (Vigodente, Rio de Janeiro, Brazil), and stored at 37 °C and 100% humidity for 30 days to allow complete curing of the sealer. The specimens were placed in silicone molds (Vigodente) to ensure that radiographs were taken in the same position, and scanned in a Brunker micro-CT device (Skyscan 1176, Bruker-micro-CT, Kontich, Belgium) with the following parameters: 70 kv, 114 mA, 14.9 mm pixel size, and rotation step of 0.3°. Images were reconstructed using NRecon V.1.6.9.4 software (Bruker micro-CT). The root canal and filling material volumes were analyzed using CTAn V.1.17.7.2 software (Bruker micro-CT). After initial scanning, the specimens were randomly divided into 2 groups of 10 mesial canals each.

### Endodontic retreatment

Filling materials were removed with PDR (25/0.06) and RECB (25/0.08) files. The same tooth was subjected to instrumentation with both systems by alternately working the two mesial canals of each tooth, resulting in 10 root canals per group. Both instruments were used in reciprocating motion and driven by a VDW motor set to “Reciproc ALL”

mode. A dental operating microscope (Alliance, São Paulo, Brazil) with 8x magnification was used to support the procedure. The files of both systems were designed for single use, so they were discarded after each use. Filling removal was considered complete when no evidence of filling material was visible on the files or on the teeth under the microscope. The re-treated specimens were then subjected to a new micro-CT scan using the same procedure as previously described.

### Supplementary root canal filling removal

The XP-Endo Finisher R file was used in both groups to remove any residual material detected during the micro-CT assessment. First, the root canals were irrigated with 5.0 mL of 2.5% NaOCl using a syringe and a 30 G needle for 30 s. After irrigation, the XP-Endo Finisher R file was operated in the root canal for 1 min at a speed of 800 rpm and a torque of 1 N.cm, performing 7 to 8 mm vertical movements along the WL. Finally, the teeth were washed with 5 mL of 17% EDTA for 15 s to remove debris, and then dried with paper points. After this supplementary step, the teeth were subjected to a third micro-CT scan.

### Statistical analysis

The quantitative data of the remaining volume (mm<sup>3</sup>) and the percentage reduction of filling material (%) obtained from the micro-CT analysis were processed using BioEstat 4.0 software. The Wilcoxon-Mann-Whitney U test was used to compare the residual volumes of filling material between the study groups, and the Wilcoxon's signed-rank test was used for pairwise comparisons within groups. The significance level was set at 5% ( $p < 0.05$ ).

## RESULTS

Descriptive statistics were performed for the remaining volume (mm<sup>3</sup>) and percent reduction of filling material. The mean values and standard deviations for each experimental group are shown in Table 1.

Both systems removed a significant amount of root canal filling material ( $p < 0.05$ ). Although the mean percentage of filling material removed was higher for A than with B PDR (91.2%) than with RECB (82.7%), no significant difference was found between the two systems after filling removal (Table 1). Similarly, the mean volumes of residual filling

| Table 1. Means and standard deviations of the quantity of remaining filling material in mm <sup>3</sup> and mean percentage of remaining filling material (%) by postoperative root instrumentations and after supplementary cleaning protocol.                                                                                                      |    |                          |                           |      |                           |                  |
|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----|--------------------------|---------------------------|------|---------------------------|------------------|
| Group                                                                                                                                                                                                                                                                                                                                                | N  | Preoperative             | Postoperative             | %    | XP-Endo Finisher R        | %                |
| ProDesign R                                                                                                                                                                                                                                                                                                                                          | 10 | 4.19 ± 1.38 <sup>a</sup> | 0.33 ± 0.20 <sup>bb</sup> | 91.2 | 0.19 ± 0.12 <sup>cc</sup> | 42.7             |
| Reciproc Blue                                                                                                                                                                                                                                                                                                                                        | 10 | 4.15 ± 1.83 <sup>a</sup> | 0.64 ± 0.54 <sup>bb</sup> | 82.7 | 0.53 ± 0.50 <sup>cc</sup> | 27.7             |
|                                                                                                                                                                                                                                                                                                                                                      |    |                          | <i>p</i> = 0.075          |      | <i>p</i> = 0.095          | <i>p</i> = 0.012 |
| The values are expressed as mean ± SD.<br>*Different lower-case letters in the mean column denote significant differences between the ProDesign R and Reciproc Blue instruments.<br>Different uppercase letters in the mean line denote significant differences between Postoperative and XP-Endo Finisher R. (Mann-Whitney U test, <i>p</i> >0.05). |    |                          |                           |      |                           |                  |

material were significantly reduced in both groups after supplementary protocol with XP-endo Finisher R (*p* < 0.05), but no statistical difference was found between groups (Table 1). All samples were found to have residual filling material in the root canals after the filling removal. Representative micro-CT reconstructions of specimens from each group are shown in Fig. 1.

DISCUSSION

Many studies have shown that reciprocating systems are effective in endodontic retreatment procedures, successfully removing most of the filling material from straight and curved canals, with performances comparable to those of continuous rotation techniques or hand files<sup>21,22</sup>. In this study, the PDR

and RECB files were found to be equally effective (91.2% and 82.7%, respectively). Therefore, the first null hypothesis tested (a) was accepted; both reciprocating instruments effectively removed most of the filling material from moderately curved canals (*p*>0.05).

The present study used de microcomputed tomography to assess the volume of filling material after the retreatment, which is the gold-standard method for root canal assessment due to its non-invasive nature, and the fact that it allows 3D analysis of several root canal anatomical parameters, both before and after treatment and retreatment<sup>4,19,23</sup>. Some similar studies assessing different reciprocating instruments used microcomputed tomography to evaluate retreatment<sup>9,21</sup>, but there are few studies on

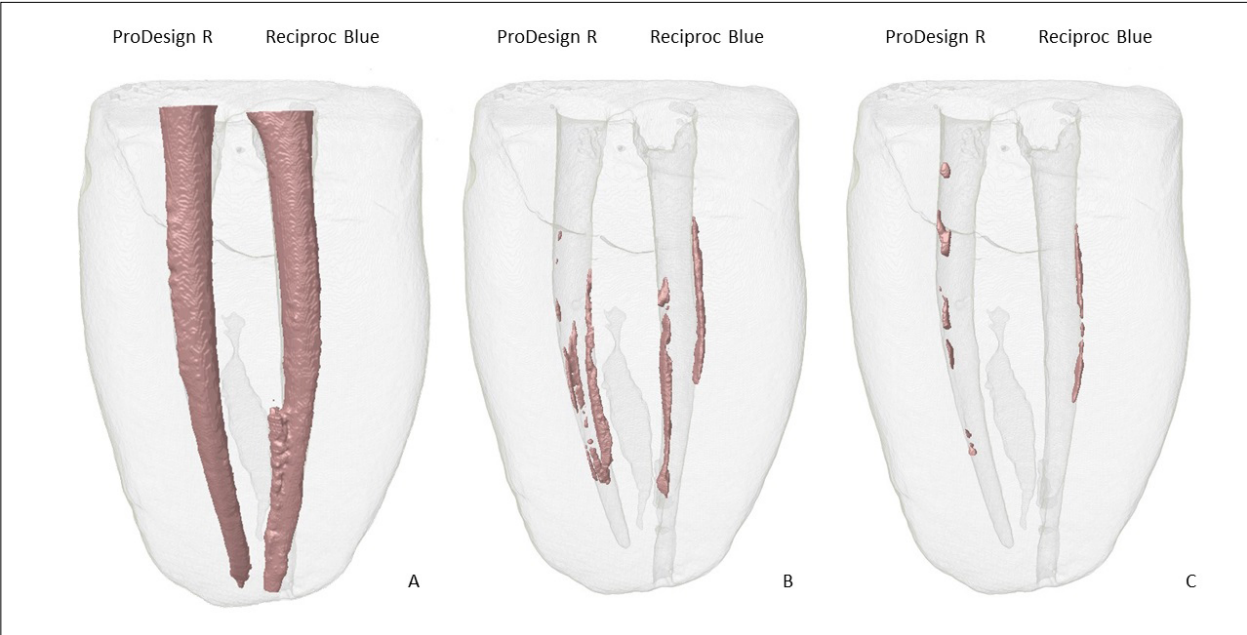


Fig. 1: A): Representative three-dimensional images showing an obturated specimen. B) After preparation with Prodesign R (PR) and Reciproc Blue (RECB) instruments. C) After supplementary cleaning protocol with XP-Endo Finisher R.

the removal of filling material by PDR and RECB files after retreatment procedures. Rodrigues et al.<sup>24</sup> tested different instruments and movements, concluding that the effectiveness of filling material removal with PDR CM files was similar to that with Reciproc R25 files with M-wire, suggesting that the type of alloy was not a relevant factor under the conditions of their study. Studies testing of RECB files have shown that their effectiveness for filling removal is comparable to that of Reciproc R25 and WaveOne Gold instruments and superior to that of manual instruments<sup>25,26</sup>. The results of the present study are consistent the literature that reports that the reciprocating instruments of the PDR and RECB systems were equally effective in removing filling material from moderately curved canals, although their thermal treatment and taper size characteristics differed from those of systems previously tested elsewhere<sup>19</sup>.

The files in both systems have S-shaped cross-sections, which may have been responsible for their similar performance during the retreatment procedure. Instrument safety is a critical factor in endodontic retreatment procedures, and instrument fractures and deformities have been described, especially in curved root canals<sup>27</sup>. The root canal curvature of the specimens and the mechanical properties of the thermally treated reciprocating instruments used in the present study may have been responsible for the single instrument fracture observed during instrumentation of the mesiobuccal canal with the PDR file.

The PDR files have a similar taper size (0.06) to RECB files (0.08) in the first 3 mm from the tip of the file, decreasing towards the cervical, with the average taper varying from 0.06 to 0.05 along the instrument, so they are likely to be subjected to stress during instrumentation, possibly resulting in a higher fracture rate. Studies have reported that an additional cleaning procedure performed with XP-endo Finisher files effectively removes filling material debris from straight and curved oval-shaped canals<sup>7,8,28</sup>. Furthermore, additional cleaning by XP-endo Finisher is useful in removing biofilm<sup>6,24</sup>, hard tissue debris<sup>10</sup>, and calcium hydroxide<sup>26</sup>.

This study also aimed to assess whether an additional finishing step could improve the cleaning process. To this end, any specimens with remaining filling, irrespective of their preparation group, underwent a finishing procedure using the XP-endo

Finisher R instrument. Another study reported the effectiveness of the XP-endo Finisher in removing residual filling materials<sup>5</sup>. The XP-endo Finisher R, specifically designed for retreatment, was found to further improve the removal of filling materials when used as an additional step in this study. The instrument's greater reach, up to 6 mm in diameter or 100 times that of a standard instrument of the same size, is attributed to the shape-memory properties and flexibility of the MaxWire alloy. This feature potentially allows the instrument to contact more dentine walls and displace filling remnants, thereby improving cleaning. These results align with a previous study reporting that XP-endo Finisher and XP-endo Finisher R instruments are equally effective in removing filling remnants<sup>27</sup>.

Although without differences between groups, in the present study, when compared the filling material removal and XP-endo Finisher R steps, it was found a significant difference, from 91.2 to 42.7% for PDR, and from 82.7 to 27.7% ( $p = 0.012$ ), showing good results in filling material remnants removal, in accordance with previous studies<sup>19,27</sup>.

Although the present study found no difference between groups, it found significant intragroup filling material removal by PDR and RECB (91.2% and 82.7%, respectively) and by XP-endo Finisher R (42.7% and 27.7% for canals treated with PDR and RECB, respectively) ( $p = 0.012$ ), in agreement with previous studies<sup>19,27</sup>.

Recently, Silva et al.<sup>7</sup> investigated the efficacy of XP-endo Finisher and XP-endo Finisher R in the removal of filling material, finding that both systems cleaned equally effectively under the conditions of their study. The results of the present study are consistent with reports that the amount of filling material observed after the use of XP-Endo Finisher R was significantly less than the amount observed before this additional cleaning. Thus, the second null hypothesis tested (b) was also rejected. This finding could be related to the austenitic memory effect of the XP-endo finisher files, which helps to extend their reach, thereby improving root canal cleanliness. Both instrumentation protocols failed to completely remove the filling material from the root canal samples in the present study, even after the additional cleaning procedure. Several studies have reported similar results, finding residual filling material in all canals, regardless of the retreatment technique used<sup>13,14,24,29</sup>.

Nevertheless, the results of the current study confirm that removing as much gutta-percha and sealer as possible by combining different instruments and complementary approaches is still an essential strategy to address the problem of cleanliness in endodontic retreatment. Overall, all samples presented remnant filling material. However, after the use of the XP-endo Finisher R instrument, the remnants were reduced to 42.7 and 27.7% for canals treated with ProDesign R and Reciproc Blue, respectively. Residual filling materials can cover areas inhabited by remaining bacteria, such as recesses, isthmuses, dentinal tubules, and ramifications<sup>30</sup>. Bacteria can remain viable in these

hard-to-reach areas protected by residual filling material, as they may not be affected by antimicrobial agents used during retreatment. The impact of these filling material remnants on root canal disinfection and, consequently, the outcome of the retreatment, is yet to be fully understood<sup>19</sup>.

## CONCLUSION

The tested systems ProDesign R and Reciproc Blue were equally effective for root canal filling removal in mesial canals of mandibular molars, but neither system completely removed the filling material. Using Xpendo finisher R as a supplementary step improved remnant removal.

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## CONFLICT OF INTERESTS

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

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# Palatal height, thickness and density according to facial biotype in Peruvian adults: a tomographic study

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## ABSTRACT

Micro-screw stability requires adequate bone quantity and quality. Micro-screws are often placed in the hard palate, in which anatomy may vary according to vertical growth. **Aim:** To compare the height, width and density of the palate according to facial biotype using cone beam computed tomography (CBCT). **Materials and Method:** Observational cross-sectional study on a sample of 39 CBCT scans of adults aged 18 to 50 years (19 females and 20 males) assigned to facial biotypes according to SN-GoGn. The height (mm), cortical width (mm) and density in Hounsfield units (HU) were measured at 20 coordinates on the left side of the palate coordinates corresponding to the combinations of four points medial to the suture (3, 5, 7 and 9 mm) and five points posterior to the incisive foramen (3, 6, 9, 12 and 15 mm). ANOVA and Kruskal-Wallis tests were used for statistical analysis at a significance level of  $p < 0.05$ . **Results:** Values were: hypodivergent (height: 2.16 - 6.32 mm; width: 1.61 - 2.02 mm; density: 1117.28 - 1182.83 HU), normodivergent (height: 2.71 - 9.21 mm; width: 1.67 - 2.08 mm; density: 1106.53 - 1177.86 HU) and hyperdivergent (height: 2.37 - 12.32 mm; width: 1.62 - 2.07 mm; density: 1088.2 - 1156.7 HU). Compared to hyperdivergent subjects, in hypodivergent individuals 60% of the measurement points had lower heights and 15% of measurement points showed thinner, denser cortices, with significant differences ( $p < 0.05$ ). **Conclusion:** Facial biotypes were found to be associated with palate bone characteristics, with greater cortical heights and widths in hyperdivergent subjects and greater densities in hypodivergent subjects.

**Keywords:** hard palate - bone density - cortical bone - cone beam computed tomography

## Altura, grosor, y densidad del paladar según biotipo facial en adultos peruanos: un estudio tomográfico

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## RESUMEN

Los microtornillos necesitan disponer de un hueso adecuado en cantidad y calidad para su estabilidad. El paladar duro es un área de colocación común de microtornillos que podría tener variaciones anatómicas debido al crecimiento vertical. **Objetivo:** Comparar la altura, el grosor y la densidad del paladar según el biotipo facial mediante tomografías computarizadas cone beam (TCCB). **Materiales y Método:** Estudio observacional transversal realizado una muestra de 39 TCCB de adultos de 18 a 50 años (19 mujeres y 20 varones) distribuidos en biotipos faciales según SN-GoGn. La altura (mm), grosor cortical (mm) y densidad en unidades Hounsfield (UH) se midieron en 20 coordenadas del lado izquierdo del paladar correspondientes a las combinaciones de cuatro puntos mediales a la sutura palatina (3, 5, 7, y 9 mm) y cinco puntos posteriores al agujero incisivo (3, 6, 9, 12, y 15 mm). Pruebas de Anova y Kruskal-Wallis fueron usadas para el análisis estadístico de la data a un nivel de significancia de  $p < 0,05$ . **Resultados:** Los valores fueron: hipodivergente (altura: 2,16 - 6,32 mm; grosor: 1,61 - 2,02 mm; densidad: 1117,28 - 1182,83 UH), normodivergente (altura: 2,71 - 9,21 mm; grosor: 1,67 - 2,08 mm; densidad: 1106,53 - 1177,86 UH) e hiperdivergente (altura: 2,37 - 12,32 mm; grosor: 1,62 - 2,07 mm; densidad: 1088,2 - 1156,7 UH). Los hipodivergentes en comparación a los hiperdivergentes tuvieron un 60% de puntos de medición con alturas inferiores y un 15% de puntos de medición con corticales adelgazadas y mayor densidad, con diferencias significativas ( $p < 0,05$ ). **Conclusión:** Los biotipos faciales se encontraron asociados a las características óseas del paladar mostrando mayores alturas y grosores corticales en hiperdivergentes y mayores densidades en hipodivergentes.

**Palabras clave:** paladar duro - densidad ósea - cortical ósea - tomografía computarizada de haz cónico.



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## INTRODUCTION

Temporary anchorage devices (TADs) are absolute anchorage systems used in orthodontics and as an alternative to maxillofacial surgery. TADs are cost-effective and require little patient collaboration<sup>1,2</sup>. Among others, the palatal region is frequently used for TAD insertion, with the paramedian and medial-anterior regions having the greatest potential due to the absence of critical neurovascular bundles<sup>3</sup>.

Maximum anchorage prevents the anchored teeth from moving<sup>4</sup>. Successful TAD placement in the palate to ensure primary stability depends largely on the amount of surrounding bone<sup>5</sup>. Therefore, before placing a TAD, it is essential to assess palatal morphology, including bone quantity and quality, in order to select the appropriate micro-screw diameter and length, and avoid the risk of penetrating and/or perforating any adjacent structures<sup>6</sup>.

To ensure micro-screw stability, a palate must have bone height  $\geq 4\text{mm}$ <sup>7</sup>, cortical width  $\geq 1\text{mm}$ <sup>8</sup>, and be sufficiently dense to prevent loosening<sup>9,10</sup>. These characteristics may differ depending on measurement towards the midline or in the anteroposterior direction, as well as other characteristics such as patient ethnicity, gender and age<sup>11,12</sup>. This leads to the need for further studies to determine the most adequate areas for TAD insertion in each population<sup>13</sup>.

Vertical facial growth is often evaluated for orthodontic purposes using cephalometric measurements<sup>10,11,14</sup>, and is closely related to morphological changes related to genetics and childhood respiratory function, with cortical bone thickness varying among different facial biotypes<sup>15-17</sup>. Therefore, the type of vertical growth should be considered when planning a palatal TAD insertion procedure<sup>10,17</sup>.

Six previous cone beam computed tomography (CBCT) studies from Brazil<sup>18</sup>, China<sup>19</sup>, Iran<sup>20</sup>, Korea<sup>21</sup>, Peru<sup>22</sup> and the United States<sup>23</sup> compared facial biotypes and palate bone characteristics, determining parameters of bone height<sup>18,19,22,23</sup>, cortical width<sup>20,22</sup> and bone density<sup>21,22</sup>. Sexual dimorphism was observed, with greater palatal height in males<sup>18</sup> and greater cortical width in females<sup>21</sup>. An association between facial biotype and palatal height<sup>19,22,23</sup> and cortical width<sup>20,22</sup> has also been found.

CBCTs are conventionally used in the orthodontic clinical setting and are useful for micro-screw

planning<sup>24,25</sup>. The palatal bone needs to be evaluated in order to determine the safest areas for insertion. The purpose of this study was to compare the height, width and density of the palate according to facial biotype using CBCT in Peruvian individuals. The null hypothesis was that there are no differences in the measurements between the different biotypes.

## MATERIALS AND METHOD

### Study design and ethics

This was a cross-sectional, analytical study developed according to the STROBE checklist. It was approved by the Institutional Research Ethics Committee of the Universidad Científica del Sur (Lima-Peru) with registration No. 189-CIEI-CIENTÍFICA-2023.

### Study sample

The sample consisted of CBCTs from 39 adults (19 females and 20 males) aged 18 to 50 years (mean age  $37.38 \pm 9.54$ ), performed at a private radiology center in Lima between 2020 and 2022. Inclusion criteria were CBCT records of patients at maximum intercuspitation, without systemic disease, genetic or congenital malformations, or long-term medication that could affect bone metabolism. CBCTs of the upper jaws showing impacted teeth, dental implants, or signs of disjunction or expansion were excluded.

### Scan selection

The data evaluated were from previous CBCT scans of patients acquired with AXR Eagle 3D equipment (Dabi Atlante, Brazil) at a UHD setting with 129 kv; 3.2–8 mA; voxel size 0.15 mm; field of view 9 cm x 9 cm; and exposure time 25 s. The CBCT images were transferred to Digital Imaging and Communication in Medicine (DICOM) format for viewing on a 14" monitor and Core i5 computer. Image segmentation was performed with OnDemand3D™ software without filters, using tools for brightness and contrast adjustment, and enlargement.

### Groups according to facial biotype

The records of the selected images included patient sex and age. The facial biotype was assessed according to Steiner's vertical cephalometric measurements with respect to the mandibular plane angle (SN-GoGn), which resulted in an average

of  $32.68 \pm 4.05$ . The sample was selected at convenience to include 13 individuals in each group of: normodivergent ( $29^\circ - 36^\circ$ ), hypodivergent ( $< 29^\circ$ ) and hyperdivergent ( $> 36^\circ$ )<sup>26</sup>.

### Pilot study and calibration

An orthodontic resident (MERR) received theoretical and practical training from a specialist in orthodontics (CITF) and calibration from an expert radiologist with more than five years' professional experience in the specialty (National Registry of Specialists N°199) for software handling and palate measurement. From a pilot study on 12 CBCTs (excluded sample), we determined intra-examiner (two weeks post-revision) and inter-examiner calibration statistics with the intraclass correlation coefficient (ICC), which were excellent for measurements of palate height (ICC: Intra-examiner  $\geq 0.999$ ; inter-examiner  $\geq 0.989$ ), cortical width (ICC: intra-examiner  $\geq 0.998$ ; inter-examiner  $\geq 0.763$ ), and density (ICC: intra-examiner  $\geq 0.985$ ; inter-examiner  $\geq 0.982$ ).

### Palatal height, width and density

All images were evaluated by the calibrated observer, who made up to six CBCT observations per day on frontal, sagittal and coronal slices. The

height (H), width (W) and density (D) values of 20 measurement points were measured. These points were established according to the anatomical structures: incisive foramen (IF), and posterior and anterior nasal spine. Palatal height was plotted between the lower and upper part of the hard palate vault (Fig. 1A). The cortical width of the palate considered only the lower area of the hard palate as the area responsible for primary stability (Fig. 1B). Palate density was measured according to the attenuation seen with the Hounsfield units (HU) indicated by the software (Fig. 1C).

### Measurement points

The initial point was located on the palatal bone adjacent to the IF, from which a perpendicular line was drawn to the horizontal plane passing through the median suture to the posterior nasal spine. A total 20 measurement points were plotted on a 4 x 5 grid on the left side of the middle of the hard palate. The intervals were marked lateral to the palatine raphe (PR) at 3, 5, 7 and 9 mm medial (M) (Fig. 1D) and at the back of the IF at 3, 6-, 9-, 12- and 15-mm posterior (P) (Fig. 1E).

### Statistical analysis

Descriptive statistics included mean and standard

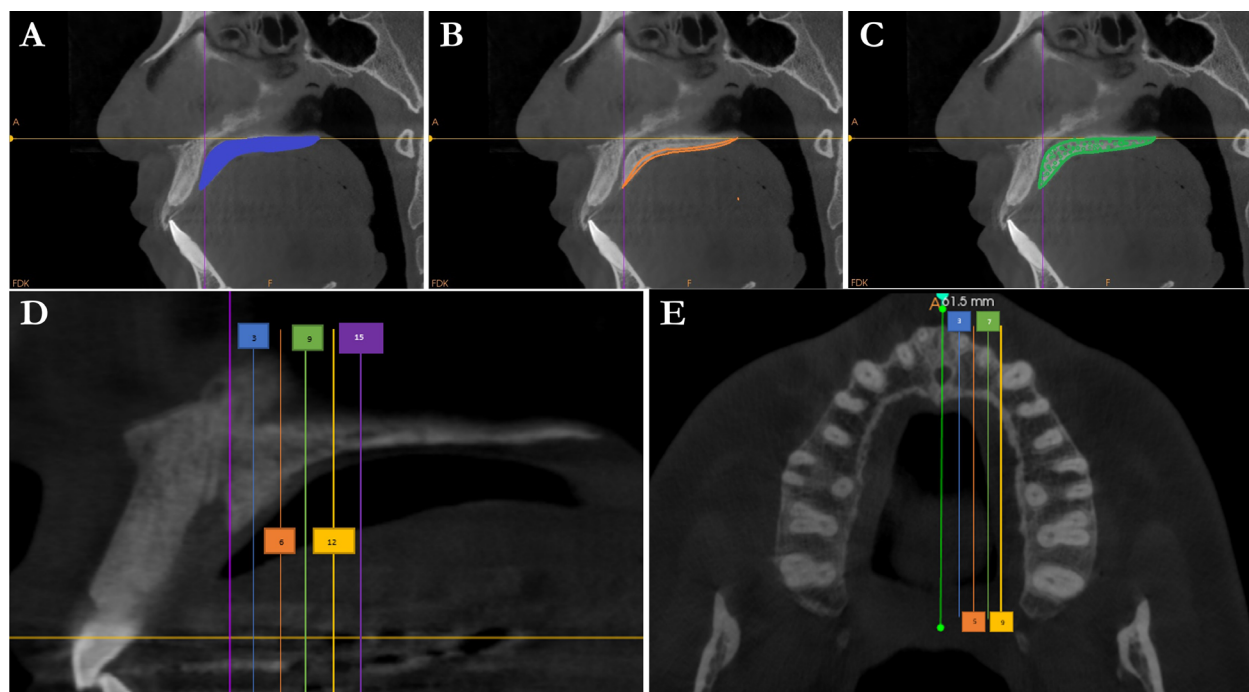


Fig. 1. Evaluation of the palatal bone in CBCT: A) height, B) cortical thickness, and C) density at the measurement points in D) cross-sectional view at 3, 6, 9, 12, and 15 mm, and E) sagittal view at 3, 5, 7, and 9 mm.

deviation (SD). Inferential ANOVA tests with Tukey's post-hoc analysis, Student's t-test for independent samples and Pearson's correlation were used when normal distribution was corroborated with the Shapiro-Wilk test. In case of non-normality, data were analyzed with the Kruskal-Wallis and Mann-Whitney U tests. Correlation was assessed as: very low (0 – 0.2), low (> 0.2 – 0.4), moderate (> 0.4 – 0.6), high (> 0.6 – 0.8) or very high (> 0.8 – 1). IBM SPSS statistical software v.26 (IBM Chicago INC) was used considering a significance level of  $p < 0.05$ .

## RESULTS

A total 2340 measurements were taken of three palatal bone characteristics (height, cortical width and density) at 20 coordinates (combinations of four medial points and five posterior points) on CBCT images of 39 patients (13 for each facial biotype). The overall mean palate height was  $5.64 \pm 3.00$  mm (range: 2.41 to 10.27 mm) (Table 1), palate cortical width was  $2.00 \pm 0.00$  mm (range: 1.64 to 2.06 mm) (Table 2) and palate density was  $1153.64 \pm 32.13$  HU (range: 1122.01 to 1158.78 HU) (Table 3).

Medial measurement of palatal height was  $M3 > M5 < M7 < M9$  and in posterior measurement it was  $P3 > P6 > P9 > P12 > P15$ . Greater heights were found in M3/9 in P3 (8.91 to 10.27 mm), while values < 6 mm were found in M5/7 in P6/9/12/15 and M3/9 in P9/12/15 (2.41 to 5.70 mm). Height was not associated with sex or age in 90% of MP ( $p < 0.05$ ), but a significant high positive correlation with SN-GoGn ( $r = 0.607$  to  $0.830$ ;  $p < 0.05$ ) was found in 70% of MP (M3/M5 at P3/6 and M7P3). The hypodivergent biotype had significantly lower heights ( $p < 0.05$ ) compared to the hyperdivergent in 60% of MP (M3/5/7 in P3/6/9 and M9 in P6/9/12) and to the normodivergent in 40% of MP (M3P3, M5/7 in P3/6/9 and M9P6) (Table 1).

Palatal bone cortical width decreased consecutively towards the lateral and posterior areas. The cortical bone was thickest at M3P3 ( $2.06 \pm 0.10$  mm) and thinnest at M9P15 ( $1.64 \pm 0.15$  mm). Width was not associated with sex and age in  $\geq 90\%$  of the MP ( $p < 0.05$ ), but was low-moderately positively correlated with SN-GoGn ( $r = 0.324$  to  $0.459$ ;  $p < 0.05$ ) in 30% of the MP (M3 in P3/6/12/15 and M5 in P3/9). The hyperdivergent biotype had significantly ( $p < 0.05$ ) greater cortical width than the hypodivergent biotype at M3P15 and M5 at P3/15 and the normodivergent

biotype at M3P15 and M9P6 (Table 2).

Palate density decreased in medial to lateral direction and in the posterior direction at P15. Density was highest at M9P12 and M3P3 (1172.46 and 1158.78 HU, respectively) and lowest at M9P15 (1122.01 HU). Density was not related to sex or age in 90% of MP ( $p < 0.05$ ) but correlated negatively low/highly significant with SN-GoGn ( $r = -0.607$  to  $-0.337$ ;  $p < 0.05$ ) in 20% of the MP (M3/5 in P3 and M3P6). The hyperdivergent biotype had lower densities ( $p < 0.05$ ) than the hypodivergent biotype at M3/5 in P3 and the normodivergent biotype at M3/5/9 in P3 (Table 3).

Figure 2 shows the comparison of biotypes according to heights (Fig. 2A), thicknesses (Fig. 2B), and densities (Fig. 2C) in the anterior-medial (M3P3 to M5P6), anterior-lateral (M7P3 to M9P6), middle-medial (M3P9 to M5P15), and middle-lateral (M7P9 to M9P15) areas of the palatal bone. The heights in all palatal areas and the thicknesses in the medial palate area were significantly greater in hyperdivergent (height: 4.27 - 9.86 mm, thickness: 1.89 - 1.99 mm) compared to hypodivergent (height: 2.91 - 6.23 mm, thickness: 1.82-1.91 mm) individuals ( $p < 0.001$  and  $p \leq 0.006$ , respectively). Densities in the medial and medio-lateral areas of the palate were significantly higher in hypodivergents (1143.04 - 1159.23 HU) compared to hyperdivergents (1119.0 - 1135.44 HU) ( $p \leq 0.049$ ).

## DISCUSSION

Treatment using micro-screws has an adequate cost-benefit ratio<sup>1-3</sup>. The palate is an area of interest for micro-screw insertion, although factors related to primary stability need to be studied<sup>10-12</sup>. Facial biotype is a biological factor related to bone growth and development, and could influence the decision-making process for deciding on the best placement site or selecting micro-screw length<sup>4,27</sup>. The results of this study show that palate bone characteristics are associated with facial biotype but not related to sex.

In the present study, palatal height correlated positively with SN-GoGn in 70% of the MP, with greater heights observed in hyperdivergent compared to hypodivergent subjects. This agrees with a previous study on posterior palate in American subjects<sup>22</sup>, but differs from studies on Chinese<sup>19</sup> and Peruvians subjects<sup>22</sup>, in which hypodivergent patients had higher anterior<sup>19,23</sup> or posterior<sup>19,22</sup> palate

**Table 1. Palatal height at the different measurement points (mm) according to age, sex, and facial biotype.**

| Parameter | Total        | Age†            | Male        | Female       | <i>p</i><br>value‡ | SN-<br>GoGn†    | Hypodivergent | Normodivergent | Hyperdivergent | <i>p</i><br>value¥ |
|-----------|--------------|-----------------|-------------|--------------|--------------------|-----------------|---------------|----------------|----------------|--------------------|
|           | Mean ± SD    | rho (p value)   | Mean ± SD   | Mean ± SD    |                    | rho (p value)   | Mean ± SD     | Mean ± SD      | Mean ± SD      |                    |
| M3P3      | 8.91 ± 3.04  | -0.016 (0.923)  | 9.24 ± 3.25 | 8.55 ± 2.85  | 0.488              | 0.830 (<0.001)* | 5.62 ± 1.61c  | 8.77 ± 1.21b   | 12.32 ± 0.96a  | <0.001*            |
| M3P6      | 6.69 ± 2.73  | -.095 (0.566)   | 7.01 ± 2.94 | 6.35 ± 2.53  | 0.652              | 0.623 (<0.001)* | 4.46 ± 1.68b  | 6.38 ± 1.28ab  | 9.22 ± 2.59a   | <0.001*            |
| M3P9      | 4.97 ± 2.11  | -0.149 (0.365)  | 5.04 ± 2.53 | 4.89 ± 1.61  | 0.632              | 0.495 (0.001)*  | 3.57 ± 1.59b  | 4.56 ± 1.62ab  | 6.59 ± 2.06a   | 0.002*             |
| M3P12     | 4.11 ± 1.85  | -0.248 (0.128)  | 4.09 ± 2.28 | 4.14 ± 1.33  | 0.938              | 0.264 (0.105)   | 3.65 ± 1.53   | 3.59 ± 1.67    | 5.10 ± 2.03    | 0.059              |
| M3P15     | 3.73 ± 1.51  | -0.184 (0.262)  | 3.50 ± 1.82 | 3.96 ± 1.10  | 0.341              | 0.193 (0.239)   | 3.45 ± 1.65   | 3.62 ± 1.49    | 4.11 ± 1.42    | 0.417              |
| M5P3      | 8.24 ± 2.93  | 0.149 (0.367)   | 8.39 ± 2.84 | 8.09 ± 3.08  | 0.761              | 0.748 (<0.001)* | 5.25 ± 2.31C  | 8.53 ± 1.20B   | 10.95 ± 1.60A  | <0.001*            |
| M5P6      | 5.44 ± 1.99  | 0.025 (0.878)   | 5.72 ± 2.25 | 5.15 ± 1.68  | 0.380              | 0.607 (<0.001)* | 3.55 ± 1.18B  | 5.85 ± 1.33A   | 6.93 ± 1.69A   | <0.001*            |
| M5P9      | 3.75 ± 1.35  | -0.142 (0.388)  | 4.04 ± 1.52 | 3.45 ± 1.09  | 0.366              | 0.392 (0.014)*  | 2.78 ± 0.71b  | 4.28 ± 1.13a   | 4.20 ± 1.55a   | 0.001*             |
| M5P12     | 2.93 ± 1.19  | -0.391 (0.014)* | 3.05 ± 1.40 | 2.81 ± 0.93  | 0.536              | 0.024 (0.883)   | 2.56 ± 0.62   | 3.34 ± 1.16    | 2.90 ± 1.55    | 0.085              |
| M5P15     | 2.64 ± 1.22  | -0.199 (0.225)  | 2.57 ± 1.55 | 2.72 ± 0.79  | 0.194              | 0.096 (0.563)   | 2.34 ± 1.09   | 2.89 ± 1.07    | 2.70 ± 1.50    | 0.480              |
| M7P3      | 8.86 ± 2.85  | 0.139 (0.399)   | 9.06 ± 3.38 | 8.64 ± 2.24  | 0.642              | 0.627 (<0.001)* | 6.32 ± 1.96b  | 9.21 ± 2.63a   | 11.05 ± 1.64a  | <0.001*            |
| M7P6      | 5.70 ± 1.71  | 0.177 (0.281)   | 5.85 ± 1.91 | 5.55 ± 1.51  | 0.594              | 0.482 (0.002)*  | 4.26 ± 1.38b  | 6.30 ± 1.16a   | 6.54 ± 1.62a   | <0.001*            |
| M7P9      | 3.84 ± 1.29  | -0.039 (0.813)  | 3.84 ± 1.45 | 3.85 ± 1.13  | 0.986              | 0.400 (0.012)*  | 2.93 ± 0.78B  | 4.20 ± 1.18A   | 4.40 ± 1.36A   | 0.004*             |
| M712      | 2.88 ± 1.10  | -0.330 (0.040)* | 3.01 ± 1.29 | 2.75 ± 0.88  | 0.390              | 0.097 (0.555)   | 2.52 ± 0.62   | 3.01 ± 1.05    | 3.13 ± 1.36    | 0.306              |
| M7P15     | 2.41 ± 1.07  | -0.086 (0.604)  | 2.42 ± 1.28 | 2.41 ± 0.82  | 0.413              | 0.019 (0.907)   | 2.16 ± 0.92   | 2.71 ± 1.00    | 2.37 ± 1.26    | 0.283              |
| M9P3      | 10.27 ± 3.44 | 0.191 (0.243)   | 9.66 ± 4.44 | 10.92 ± 1.81 | 0.517              | 0.434 (0.006)*  | 8.62 ± 3.24   | 10.78 ± 4.31   | 11.42 ± 1.92   | 0.091              |
| M9P6      | 7.45 ± 2.43  | 0.165 (0.315)   | 7.42 ± 2.94 | 7.47 ± 1.82  | 0.800              | 0.510 (0.001)*  | 5.70 ± 2.15B  | 7.92 ± 1.98A   | 8.72 ± 2.08A   | 0.002*             |
| M9P9      | 5.12 ± 1.98  | 0.050 (0.762)   | 5.25 ± 2.45 | 4.97 ± 1.37  | 0.593              | 0.430 (0.006)*  | 3.87 ± 1.50B  | 5.36 ± 1.96AB  | 6.12 ± 1.86A   | 0.009*             |
| M9P12     | 4.19 ± 1.79  | 0.009 (0.956)   | 4.30 ± 2.17 | 4.08 ± 1.33  | 0.712              | 0.387 (0.015)*  | 3.12 ± 0.97B  | 5.28 ± 1.61AB  | 5.18 ± 2.08A   | 0.010*             |
| M9P15     | 3.78 ± 1.91  | -0.039 (0.812)  | 3.96 ± 2.53 | 3.59 ± 0.92  | 0.546              | 0.350 (0.029)*  | 2.85 ± 1.15   | 3.98 ± 1.98    | 4.51 ± 2.18    | 0.075              |

SD: standard deviation. †Pearson Correlation Test. ‡Independent samples Student's T-test or Mann-Whitney U test. ¥ ANOVA test with Tukey post-hoc or Kruskal-Wallis test. \**p* < 0.05

heights close to the PR compared to other biotypes, while another study on Brazilian subjects<sup>18</sup> found no differences. The measurement of heights in distant areas using the perpendicular to the palatal plane as a reference may not be representative due to the domed shape of the palate<sup>4,27</sup>.

The insertion of mini-screws requires a palatal

height of  $\geq 4$  mm<sup>7</sup>. In this study, this required palatal height was observed from anterolateral up to 9 mm posterolateral in hyperdivergent (4.2 - 12.3 mm) and normodivergent subjects (4.2 - 10.8 mm) and up to 6 mm posterolateral (except M5P6) in hypodivergent subjects (4.3 - 8.6 mm). Clinical comparison with previous studies showed similar results in

**Table 2. Palatal cortical thickness (mm) at the different measurement points according to age, sex, and facial biotype.**

| Parameter | Total<br>Mean ± SD | Age†<br>r (p valor) | Male<br>Mean ± SD | Female<br>Mean ± SD | p value‡ | SN-GoGn†<br>r (p value) | Hypodivergent<br>Mean ± SD | Normodivergent<br>Mean ± SD | Hyperdivergent<br>Mean ± SD | p value¥ |
|-----------|--------------------|---------------------|-------------------|---------------------|----------|-------------------------|----------------------------|-----------------------------|-----------------------------|----------|
| M3P3      | 2.06 ± 0.10        | 0.155 (0.346)       | 2.06 ± 0.12       | 2.05 ± 0.04         | 0.042*   | 0.367 (0.021)*          | 2.02 ± 0.04                | 2.08 ± 0.13                 | 2.07 ± 0.08                 | 0.076    |
| M3P6      | 1.96 ± 0.10        | -0.110 (0.504)      | 1.95 ± 0.11       | 1.97 ± 0.09         | 0.209    | 0.324 (0.044)*          | 1.94 ± 0.09                | 1.95 ± 0.09                 | 1.99 ± 0.11                 | 0.117    |
| M3P9      | 1.95 ± 0.08        | -0.095 (0.566)      | 1.95 ± 0.09       | 1.95 ± 0.08         | 0.868    | 0.273 (0.093)           | 1.93 ± 0.06                | 1.94 ± 0.10                 | 1.98 ± 0.09                 | 0.093    |
| M3P12     | 1.89 ± 0.09        | -0.119 (0.469)      | 1.88 ± 0.10       | 1.91 ± 0.08         | 0.360    | 0.444 (0.005)*          | 1.86 ± 0.06                | 1.88 ± 0.11                 | 1.94 ± 0.08                 | 0.052    |
| M3P15     | 1.85 ± 0.09        | -0.078 (0.635)      | 1.84 ± 0.09       | 1.87 ± 0.08         | 0.253    | 0.394 (0.013)*          | 1.82 ± 0.05b               | 1.81 ± 0.07b                | 1.91 ± 0.10a                | 0.005*   |
| M5P3      | 1.90 ± 0.13        | -0.119 (0.471)      | 1.90 ± 0.13       | 1.91 ± 0.14         | 0.774    | 0.459 (0.003)*          | 1.82 ± 0.13b               | 1.89 ± 0.10ab               | 1.99 ± 0.12a                | 0.007*   |
| M5P6      | 1.88 ± 0.12        | -0.317 (0.049)*     | 1.89 ± 0.11       | 1.86 ± 0.12         | 0.181    | 0.023 (0.888)           | 1.86 ± 0.13                | 1.86 ± 0.09                 | 1.90 ± 0.13                 | 0.774    |
| M5P9      | 1.84 ± 0.11        | -0.165 (0.315)      | 1.84 ± 0.12       | 1.85 ± 0.09         | 0.892    | 0.374 (0.019)*          | 1.80 ± 0.14                | 1.85 ± 0.08                 | 1.88 ± 0.08                 | 0.054    |
| M5P12     | 1.83 ± 0.12        | 0.125 (0.448)       | 1.83 ± 0.14       | 1.82 ± 0.10         | 0.582    | 0.298 (0.066)           | 1.77 ± 0.14                | 1.84 ± 0.06                 | 1.87 ± 0.13                 | 0.109    |
| M5P15     | 1.79 ± 0.08        | 0.141 (0.392)       | 1.78 ± 0.09       | 1.79 ± 0.07         | 0.556    | 0.013 (0.937)           | 1.75 ± 0.10B               | 1.83 ± 0.08A                | 1.78 ± 0.05AB               | 0.044*   |
| M7P3      | 1.84 ± 0.10        | -0.161 (0.328)      | 1.86 ± 0.10       | 1.81 ± 0.09         | 0.330    | -0.076 (0.645)          | 1.85 ± 0.10                | 1.85 ± 0.11                 | 1.82 ± 0.08                 | 0.941    |
| M7P6      | 1.79 ± 0.11        | 0.186 (0.258)       | 1.81 ± 0.09       | 1.76 ± 0.13         | 0.414    | -0.266 (0.102)          | 1.83 ± 0.10                | 1.78 ± 0.12                 | 1.75 ± 0.10                 | 0.201    |
| M7P9      | 1.74 ± 0.12        | 0.280 (0.084)       | 1.79 ± 0.11       | 1.69 ± 0.10         | 0.030*   | -0.198 (0.226)          | 1.76 ± 0.14                | 1.75 ± 0.09                 | 1.71 ± 0.11                 | 0.520    |
| M7P12     | 1.73 ± 0.09        | 0.206 (0.209)       | 1.74 ± 0.10       | 1.72 ± 0.07         | 0.398    | -0.251 (0.123)          | 1.74 ± 0.12                | 1.74 ± 0.07                 | 1.71 ± 0.06                 | 0.550    |
| M7P15     | 1.70 ± 0.09        | -0.129 (0.435)      | 1.73 ± 0.07       | 1.68 ± 0.10         | 0.220    | -0.169 (0.305)          | 1.72 ± 0.10                | 1.69 ± 0.08                 | 1.70 ± 0.08                 | 0.396    |
| M9P3      | 1.86 ± 0.12        | -0.214 (0.191)      | 1.87 ± 0.13       | 1.85 ± 0.12         | 0.630    | -0.044 (0.793)          | 1.87 ± 0.13                | 1.83 ± 0.11                 | 1.89 ± 0.13                 | 0.369    |
| M9P6      | 1.75 ± 0.07        | -0.175 (0.286)      | 1.75 ± 0.05       | 1.74 ± 0.08         | 0.282    | 0.005 (0.977)           | 1.75 ± 0.06ab              | 1.71 ± 0.05b                | 1.78 ± 0.07a                | 0.033*   |
| M9P9      | 1.69 ± 0.09        | -0.096 (0.561)      | 1.70 ± 0.07       | 1.67 ± 0.11         | 0.227    | 0.132 (0.422)           | 1.68 ± 0.08                | 1.70 ± 0.08                 | 1.68 ± 0.11                 | 0.863    |
| M9P12     | 1.70 ± 0.06        | -0.167 (0.311)      | 1.71 ± 0.06       | 1.69 ± 0.07         | 0.398    | -0.214 (0.192)          | 1.71 ± 0.05                | 1.71 ± 0.06                 | 1.67 ± 0.07                 | 0.157    |
| M9P15     | 1.64 ± 0.15        | 0.148 (0.370)       | 1.66 ± 0.09       | 1.61 ± 0.19         | 0.683    | 0.140 (0.395)           | 1.61 ± 0.12                | 1.67 ± 0.10                 | 1.62 ± 0.21                 | 0.678    |

SD: standard deviation. †Pearson Correlation Test. ‡Independent samples Student's T-test or Mann-Whitney U test. ¥ ANOVA test with Tukey post-hoc or Kruskal-Wallis test. \*p < 0.05

hyperdivergent individuals (China<sup>19</sup>: 4.4 - 7.5 mm; Peru<sup>22</sup>: 7 - 12 mm; Brazil<sup>18</sup>: 6.2 - 12.8 mm), but with variations with respect to normodivergent (≈China<sup>19</sup>: 4.9 - 8.6 mm; ≈Peru<sup>22</sup>: 5.8 - 10.1 mm; ↑Brazil<sup>18</sup>: 6.8 - 13 mm) and hypodivergent subjects (≈China<sup>19</sup>: 4.8 - 9.4 mm; ↑Peru<sup>22</sup>: 8 - 11.8 mm; ↑Brazil<sup>18</sup>: 6.5 -

12 mm). It was of note that, as in the present study, some of the previous studies<sup>19,22</sup> did not control for open bite, which according to studies from Brazil<sup>18</sup> and Thailand<sup>27</sup>, influences palatal height, possibly due to a dentoalveolar compensatory effect in the long-face pattern<sup>4</sup>.

**Table 3. Palatal density (HU) at the different measurement points according to age, sex, and facial biotype.**

| Parameter | Total           | Age†            | Male            | Female          | p value‡ | SN-GoGn†         | Hypodivergent     | Normodivergent   | Hyperdivergent    | p value¥ |
|-----------|-----------------|-----------------|-----------------|-----------------|----------|------------------|-------------------|------------------|-------------------|----------|
|           | Mean ± SD       | r (p value)     | Mean ± SD       | Mean ± SD       |          | r (p value)      | Mean ± SD         | Mean ± SD        | Mean ± SD         |          |
| M3P3      | 1158.78 ± 64.17 | 0.337 (0.036)*  | 1162.15 ± 62.72 | 1155.22 ± 67.19 | 0.741    | -0.337 (0.036)*  | 1182.01 ± 83.85   | 1148.41 ± 58.78  | 1145.91 ± 40.92   | 0.193    |
| M3P6      | 1130.02 ± 55.75 | 0.025 (0.881)   | 1136.17 ± 40.97 | 1123.54 ± 68.58 | 0.725    | -0.607 (<0.001)* | 1164.13 ± 36.87A  | 1137.72 ± 58.52A | 1088.20 ± 42.89B  | 0.001*   |
| M3P9      | 1143.57 ± 67.13 | 0.257 (0.114)   | 1125.30 ± 66.65 | 1162.81 ± 63.74 | 0.081    | -0.158 (0.336)   | 1160.78 ± 55.73   | 1127.40 ± 90.27  | 1142.53 ± 49.10   | 0.533    |
| M3P12     | 1126.13 ± 52.47 | 0.015 (0.929)   | 1111.03 ± 62.21 | 1142.03 ± 34.73 | 0.063    | -0.269 (0.098)   | 1145.16 ± 47.63   | 1112.88 ± 58.49  | 1120.36 ± 49.09   | 0.266    |
| M3P15     | 1145.63 ± 48.96 | 0.276 (0.089)   | 1132.64 ± 49.66 | 1159.30 ± 45.52 | 0.014    | -0.029 (0.860)   | 1137.12 ± 46.80   | 1160.63 ± 42.95  | 1139.13 ± 56.45   | 0.405    |
| M5P3      | 1138.69 ± 51.94 | -0.064 (0.698)  | 1128.29 ± 42.45 | 1149.65 ± 59.56 | 0.203    | -0.377 (0.018)*  | 1151.03 ± 43.73a  | 1163.65 ± 37.95a | 1101.40 ± 53.34b  | 0.005*   |
| M5P6      | 1141.84 ± 44.20 | 0.172 (0.295)   | 1137.16 ± 47.46 | 1146.76 ± 41.18 | 0.877    | -0.048 (0.772)   | 1139.77 ± 39.14   | 1145.25 ± 57.38  | 1140.49 ± 36.62   | 0.945    |
| M5P9      | 1135.06 ± 48.54 | 0.139 (0.400)   | 1126.52 ± 52.27 | 1144.06 ± 43.86 | 0.265    | 0.004 (0.980)    | 1117.46 ± 42.25B  | 1162.05 ± 50.62A | 1125.68 ± 43.53AB | 0.040*   |
| M5P12     | 1132.86 ± 65.03 | -0.114 (0.489)  | 1121.62 ± 57.50 | 1144.69 ± 71.76 | 0.332    | -0.278 (0.086)   | 1161.79 ± 67.74   | 1135.35 ± 62.32  | 1101.43 ± 54.17   | 0.056    |
| M5P15     | 1129.96 ± 62.63 | -0.158 (0.336)  | 1134.08 ± 71.73 | 1125.61 ± 53.02 | 0.704    | -0.126 (0.444)   | 1135.96 ± 74.25   | 1134.78 ± 69.23  | 1119.14 ± 43.87   | 0.538    |
| M7P3      | 1133.99 ± 55.53 | -0.104 (0.530)  | 1139.36 ± 53.55 | 1128.34 ± 58.45 | 0.543    | -0.024 (0.883)   | 1122.00 ± 56.54   | 1135.56 ± 50.14  | 1144.41 ± 61.46   | 0.891    |
| M7P6      | 1134.15 ± 39.95 | 0.211 (0.196)   | 1139.41 ± 41.92 | 1128.62 ± 38.09 | 0.406    | 0.080 (0.630)    | 1135.80 ± 36.68   | 1119.55 ± 37.16  | 1147.10 ± 43.73   | 0.213    |
| M7P9      | 1146.45 ± 54.46 | -0.231 (0.158)  | 1151.94 ± 63.65 | 1140.67 ± 43.78 | 0.482    | -0.265 (0.103)   | 1164.12 ± 65.57   | 1126.40 ± 51.50  | 1148.82 ± 40.61   | 0.210    |
| M7P12     | 1136.37 ± 49.32 | -0.394 (0.013)* | 1134.49 ± 50.12 | 1138.34 ± 49.76 | 0.978    | -0.155 (0.348)   | 1142.11 ± 29.86   | 1131.24 ± 57.48  | 1135.74 ± 58.68   | 0.349    |
| M7P15     | 1132.23 ± 51.09 | 0.109 (0.510)   | 1135.08 ± 47.08 | 1129.22 ± 56.14 | 0.725    | -0.077 (0.641)   | 1137.29 ± 49.92   | 1120.62 ± 63.26  | 1138.77 ± 39.38   | 0.615    |
| M9P3      | 1128.38 ± 59.73 | -0.055 (0.741)  | 1108.97 ± 52.86 | 1148.81 ± 61.03 | 0.099    | -0.103 (0.532)   | 1117.28 ± 44.96ab | 1172.74 ± 58.19a | 1095.12 ± 49.28b  | 0.010*   |
| M9P6      | 1127.87 ± 50.82 | 0.071 (0.669)   | 1127.56 ± 62.68 | 1128.20 ± 36.12 | 0.969    | -0.213 (0.193)   | 1133.12 ± 40.05   | 1139.79 ± 44.04  | 1110.71 ± 64.28   | 0.245    |
| M9P9      | 1128.39 ± 45.73 | 0.002 (0.990)   | 1122.55 ± 31.30 | 1134.54 ± 57.46 | 0.421    | -0.030 (0.858)   | 1124.35 ± 55.19   | 1139.35 ± 47.10  | 1121.47 ± 34.01   | 0.576    |
| M912      | 1172.46 ± 52.30 | -0.140 (0.395)  | 1158.54 ± 48.10 | 1187.12 ± 53.76 | 0.088    | -0.177 (0.280)   | 1182.83 ± 50.18   | 1177.86 ± 57.05  | 1156.70 ± 49.72   | 0.411    |
| M9P15     | 1122.01 ± 48.26 | -0.173 (0.292)  | 1131.66 ± 37.23 | 1111.85 ± 56.94 | 0.054    | -0.316 (0.050)*  | 1148.37 ± 35.28   | 1106.53 ± 44.99  | 1111.13 ± 54.47   | 0.050    |

SD: standard deviation. †Pearson Correlation Test. ‡Independent samples Student's T-test or Mann-Whitney U test. ¥ANOVA test with Tukey post-hoc or Kruskal-Wallis test. \*p < 0.05

In the current study, palatal cortical width correlated positively with SN-GoGn in 30% of the MP with greater width in hyperdivergent compared to hypodivergent subjects. This result contradicts findings by Iranian<sup>20</sup> and Peruvian studies<sup>22</sup> in which the width was greater in hypodivergent > hyperdivergent > normodivergent subjects in lateral

areas close to the PR. Our study supports differences with the previous studies in the mapping of palate measurement towards the posterior of the IF, which showed greater distance in mm between each lateral and posterior MP (4 - 3 - 4, respectively)<sup>20,22</sup>, as opposed to the present study (3 - 2 - 3, respectively). Cortical widths ≥1 mm, which provide better

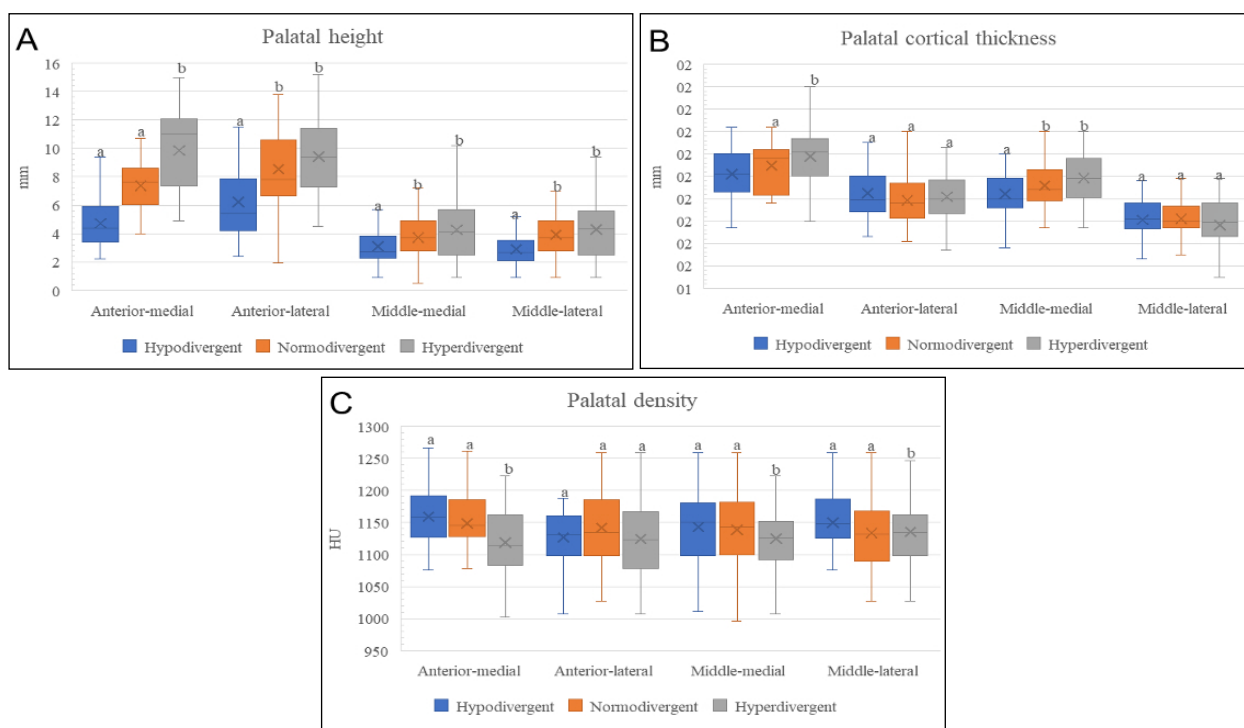


Fig. 2. Comparison of the palatal bone areas for: A) height, B) cortical thickness and C) density according to biotype. Different letters indicate significant differences with Kruskal-Wallis test.  $p < 0.05$ .

micro-screw anchorage<sup>8</sup>, were found in the total measurements of all three biotypes (1.6 to 2.1 mm). However, more areas of the palate had greater heights in the hyperdivergent and normodivergent biotypes, at 5 mm lateral and 12 mm posterior, than in the hypodivergent biotypes, at 3 mm lateral and 12 mm posterior. There were clinical differences with a study reporting a smaller width in Iranians<sup>20</sup> (0.7 - 1.7 mm), being greater in hypodivergent and normodivergent (up to 1.6 mm) than hyperdivergent biotypes (up to 1.4 mm) at 3 mm lateral and 16 mm posterior, while another study on Peruvians<sup>22</sup> reported greater width (1.1 - 3.0 mm) in hypodivergent (up to 3.0 mm), followed by hyperdivergent (up to 2.6 mm) and normodivergent subjects (up to 2.2 mm). These data are of interest in the mechanics of the micro-screws for selecting the most adequate thread types according to increased cortical width<sup>28</sup>. Palate density in this study correlated negatively with SN-GoGn in 20% of the MP, being higher in hypodivergent and normodivergent than in hyperdivergent biotypes. Differences in densities could be influenced by masticatory muscle development and masticatory functional load<sup>12,23</sup>. However, other studies on Korean<sup>21</sup> and Peruvian<sup>22</sup> populations found no differences between facial

biotypes. Although Hounsfield units are often used to assess bone quality for micro-screw placement, there are no clinically established cut-off points<sup>29</sup>. There are various methodological differences between the current study and previous ones, such as: (a) inclusion of other ethnicities<sup>18-21,23</sup>, b) non-proportional distribution by sex<sup>20,22</sup> or facial biotype<sup>18-20</sup>, c) younger subject ages including youths aged 10 to  $\leq 21$  years<sup>21</sup>, adults aged 18 to  $\leq 35$  years<sup>19,22,23</sup> or age not reported<sup>18,20</sup>, d) CBCT with voxel size  $> 0.3 - 0.6$  mm<sup>18-20,22,23</sup> or not reported<sup>21</sup>, e) different biotype analysis with SN-GoMe<sup>19,20,22</sup>, facial height index<sup>22</sup> and mandibular plane angle<sup>21,23</sup>, f) different reference MP based on teeth<sup>18,23</sup> and g) different bone density analysis with attenuation coefficients.<sup>22</sup>

This study evaluated three palatal bone features that are clinically relevant for determining adequate micro-screw placement, including a sample with evenly distributed sex and biotype, and analyzed a large area of palate with small voxel CBCT. However, the sample size was limited, so the results must be interpreted with caution. Studies with a larger sample size are recommended to increase the possibility of generalizing the results.

## CONCLUSION

Within the limitations of this study, it was concluded that the facial biotype assessed with the SN-GoGn angle is associated with palate bone characteristics, regardless of sex or age. Hyperdivergent patients

had greater heights in the anterior area and thicker cortices in the paramedial area, while hypodivergent patients had greater densities in the anteromedial area of the palate.

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## CONFLICT OF INTERESTS

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

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# Upregulation of cannabinoid receptor gene expression in oral tissues subjected to hyposalivation and periodontitis

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## ABSTRACT

Cannabinoid receptors (CB<sub>1</sub> and 2 (CB<sub>1</sub>r and CB<sub>2</sub>r) are present in periodontal tissues and salivary glands and play a significant role in the pathogenesis of oral diseases. **Aim:** To study the gene expression of CB<sub>1</sub>r and CB<sub>2</sub>r in gingival tissue and submandibular glands (SMG) of rats subjected to experimental periodontitis (EP) and/or hyposalivation (H), and discuss their possible implication in oral health. **Material and methods:** Rats were divided into controls, rats with EP induced by application of lipopolysaccharide (1 mg/ml), rats with H induced by bilateral submandibulectomy, and rats with both EP and H. Alveolar bone loss was measured with a digital caliper, mRNA expression of CB<sub>1</sub>r and bone parameters were determined by endpoint RT-PCR, PGE<sub>2</sub> glandular content was assessed by radioimmunoassay, and stimulated salivary secretion was collected and weighed. **Results:** CB<sub>1</sub>r and CB<sub>2</sub>r mRNA in gingival tissue were upregulated in groups EP, H and EP+H as compared to controls (CB<sub>1</sub>r (AU): C 0.27±0.06<sup>A</sup>, EP 0.46±0.05<sup>B</sup>, H 0.51±0.08<sup>B</sup>, EP+H 0.49±0.1<sup>B</sup>; CB<sub>2</sub>r (AU): C 0.38±0.04<sup>A</sup>, EP 1.22±0.08<sup>B</sup>, H 0.98±0.06<sup>C</sup>, EP+H 1.041±0.1B<sup>C</sup>). Groups EP, H and EP+H showed alveolar bone loss as opposed to controls, while only groups EP and EP+H evidenced RANKL/OPG imbalance measured in the dental attachment tissue. Likewise, CB<sub>1</sub>r and CB<sub>2</sub>r mRNA in SMG showed upregulation in group EP as compared to controls (CB<sub>1</sub>r (AU): C 0.52±0.09<sup>A</sup>, EP 0.95±0.13<sup>B</sup>; CB<sub>2</sub> (AU): C 0.23±0.12<sup>A</sup>, EP 1.48±0.22<sup>B</sup>). In group EP, salivary secretion was lower and the content of PGE<sub>2</sub> (inhibitory mediator of said function) was higher than in controls. **Conclusion:** The upregulation of CB<sub>2</sub>r in oral tissues under the pathophysiological conditions studied suggests that they participate actively in the response to oral diseases.

**Keywords:** cannabinoid receptors - submandibular gland excision - lipopolysaccharides - periodontium - inflammation.

## Regulación positiva de la expresión génica de los receptores cannabinoides en los tejidos bucales sometidos a hiposalivación y periodontitis

### RESUMEN

Los receptores cannabinoides (CB<sub>1</sub> y 2 (CB<sub>1</sub>r y CB<sub>2</sub>r) están presentes en los tejidos periodontales y en las glándulas salivales, y desempeñan un papel importante en la patogénesis de las enfermedades orales. **Objetivo:** Estudiar la expresión génica de CB<sub>1</sub>r y CB<sub>2</sub>r en tejido gingival y glándulas submandibulares (GSM) de ratas sometidas a periodontitis experimental (PE) y/o hiposalivación(H) y discutir su posible implicancia en la salud oral. **Materiales y método:** Las ratas se dividieron en controles, ratas con PE inducida por aplicación de lipopolisacáridos (1 mg/ml, grupo LPS), ratas con H inducida por submandibulectomía bilateral y ratas con EP y H. Se evaluó la pérdida ósea alveolar mediante un calibre digital, se determinó la expresión de ARNm de CB<sub>1</sub>r y los parámetros óseos mediante RT-PCR de punto final, se evaluó el contenido glandular de PGE<sub>2</sub> mediante radioinmunoanálisis y se recogió y pesó la secreción salival estimulada. **Resultados:** el ARNm de CB<sub>1</sub>r y CB<sub>2</sub>r en el tejido gingival mostró regulación positiva en los grupos EP, H y EP+H en comparación con los controles (CB<sub>1</sub>r (UA): C 0,27±0,06A, EP 0,46±0,05<sup>B</sup>, H 0,51±0,08<sup>B</sup>, EP+H 0,49±0,1<sup>B</sup>; CB<sub>2</sub>r (UA): C 0,38±0,04<sup>A</sup>, EP 1,22±0,08<sup>B</sup>, 0,98±0,06<sup>C</sup>, EP+H 1,041±0,1B<sup>C</sup>). Los grupos EP, H y EP+H mostraron pérdida ósea alveolar en comparación con los controles, mientras que sólo los grupos EP y EP+H evidenciaron un desequilibrio RANKL/OPG medido en el tejido de inserción dentaria. Asimismo, los ARNm de CB<sub>1</sub>r y CB<sub>2</sub>r en la GSM mostraron regulación positiva en el grupo EP en comparación con el grupo control (CB<sub>1</sub>r (UA): C 0,52±0,09<sup>A</sup>, EP 0,95±0,13<sup>B</sup>; CB<sub>2</sub> (UA): C 0,23±0,12<sup>A</sup>, EP 1,48±0,22<sup>B</sup>). En el grupo EP, la secreción salival fue menor y el contenido de PGE<sub>2</sub> (mediador inhibidor de dicha función) aumentó en comparación con el grupo control. **Conclusión:** la regulación positiva de los CB<sub>2</sub>r en los tejidos orales en las condiciones fisiopatológicas estudiadas sugiere su participación activa en la respuesta a enfermedades de la cavidad bucal.

**Palabras clave:** receptores cannabinoides - submandibulectomía-lipopolisacáridos - periodonto - inflamación

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## INTRODUCTION

Cannabinoid (CB) receptors 1 and 2 (CB1r and CB2r) are central elements of the endocannabinoid system (ECS). They are both involved in a range of signaling pathways that influence physiological processes. CB1r is primarily present in the central nervous system (CNS) and influences neurotransmission, while CB2r is mainly present in peripheral tissues, participating in the modulation of immune and inflammatory responses<sup>1</sup>. The signaling mechanisms of both receptors contribute to the wide range of effects attributed to cannabinoids. CBrs are activated by endocannabinoids such as anandamide and 2-arachidonoylglycerol, as well as by exogenous cannabinoids such as  $\Delta$ -9-Tetrahydrocannabinol. These receptors are primarily coupled with Gi/o proteins, which inhibit adenylyl cyclase enzyme and, subsequently, affect various downstream cellular processes. They also promote MAPK activity, thus influencing cellular processes, including gene expression, cell proliferation, and survival. Only CB1r has been shown to signal via ion channels. It inhibits voltage-gated calcium channels, reducing calcium influx, and activates inwardly rectifying potassium channels, causing hyperpolarization. CB1r and CB2r also influence other second messenger systems such as phosphatidyl inositol 3-OH kinase (PI3K)-Akt pathway, through which CB1r has a role in cell survival and apoptosis, whereas CB2r takes part in cell survival, migration and immune responses. Extracellular signal-regulated kinase (ERK) is another pathway activated by CB1r, becoming involved in cellular growth and differentiation. Furthermore, NF- $\kappa$ B and JAK/STAT pathways can be activated by CB2r, playing a role in immune and inflammatory responses<sup>1,2</sup>.

CBrs are present in periodontal tissues and play a significant role in the pathogenesis of oral diseases<sup>3</sup>. Periodontitis is a chronic inflammatory disease generated mainly by the accumulation of dental biofilm, inflammation of the dental insertion tissues and formation of periodontal pockets. It causes the resorption of alveolar bone and the loss of insertion periodontal tissue, and can even lead to tooth loss if untreated<sup>4</sup>. Recent studies have demonstrated the involvement of the ECS in the modulation of immune responses in periodontitis<sup>5-7</sup>. Activation of CB2r exerts anti-inflammatory effects, bringing the potential capability to reduce the severity of inflammation and tissue destruction associated with

the disease<sup>8</sup>. Destruction of periodontal tissues results from the release of harmful molecules by periodontopathogenic bacteria and host-derived proinflammatory mediators released during the immune response<sup>4,9</sup>. The inflammatory environment leads to an imbalance between alveolar bone resorption and formation, favoring the former<sup>10</sup>. Bone metabolism is regulated by the receptor activator of nuclear factor-kappa B (RANK), its ligand (RANKL) and osteoprotegerin (OPG), a decoy receptor<sup>11,12</sup>. Thus, controlling inflammation by CBrs activation could be a useful tool for avoiding bone metabolism imbalance, as reported in preclinical studies<sup>13</sup>.

Cannabinoid receptors are also present in salivary glands. In addition to their role in inflammation, CBrs are involved in the regulation of saliva production<sup>14,15</sup>. Saliva is composed of water as solvent and electrolytes, proteins and other molecules as solutes. It is crucial in the mouth to maintain the integrity of the mucous membranes and hard tissues<sup>16-18</sup>. Reduction in saliva production affects the integrity of oral structures, increasing the risk of infection by microorganisms and the development of dental caries<sup>19</sup>. Furthermore, the submandibular gland has been shown to play an immunomodulatory role, controlling oral homeostasis<sup>20</sup>. Hyposalivation (H) and the subjective sensation of dry mouth, called xerostomia, are health problems that affect millions of people around the world<sup>21,22</sup>.

Periodontitis-induced hyposalivation, or reduced saliva flow, can exacerbate oral health problems by impairing the natural cleansing mechanisms of the oral cavity and increasing the risk of infection. Studies have indicated that activation of CBrs in gingival tissues and submandibular glands can differentially influence salivary gland function, which could lead to changes in saliva secretion<sup>14,23,24</sup>. Furthermore, the anti-inflammatory effect induced by CB2r activation may also help to alleviate the inflammation of oral tissues, thus improving saliva production<sup>24</sup>. This dual role of cannabinoid receptors in both inflammation and salivary gland function highlights their potential as therapeutic targets for managing periodontal disease and its associated complications, such as hyposalivation.

In line with these findings, an increasing number of studies have highlighted the therapeutic potential of cannabinoid-based interventions for various oral

pathologies. Evidence from preclinical and clinical research supports the use of cannabinoids for their anti-inflammatory, analgesic, antimicrobial, and immunomodulatory properties in conditions such as periodontitis, oral mucositis, temporomandibular joint disorders, and even oral cancer<sup>6,25,26</sup>. These studies underscore the relevance of the endocannabinoid system as a promising target in oral medicine, paving the way for the development of novel cannabinoid-based therapeutic strategies aimed at improving oral health outcomes.

In a recently published article, using the same experimental model as in the current study, we showed an increase in gingival inflammatory parameters and bone loss in rats subjected to hyposalivation (H) compared to controls. Furthermore, we observed higher levels of inflammatory mediators in the experimental periodontitis (EP) group than in the H group, resulting in greater damage to periodontal tissues. However, we concluded that concomitant exposure to EP and H did not show clear aggregate effects<sup>27</sup>. The aim of the current work was to study the gene expression of CB1r and CB2r in periodontal tissue and submandibular glands of rats subjected to experimental periodontitis and/or hyposalivation, and to discuss their possible implications for oral health.

## MATERIALS AND METHODS

### Animals

Adult male Wistar rats (weighing 300-350 g) from the laboratory's own colony were housed in group cages in a controlled environment with a 12-hour light/dark cycle (0800-2000). Room temperature was kept at 22±2°C, and the animals had unlimited access to standard rat food and tap water. The experimental protocols were approved by the Animal Care Committee of the Dental School at the University of Buenos Aires, Argentina (CICUAL-ODON/FOUBA N° 013/2016), and adhered to the European Communities Council Directive 2010/63/UE. All animal experiments followed the ARRIVE guidelines 2.0.

### Design

Twenty-four rats were divided into four groups of six: 1) control rats, 2) rats subjected to experimental periodontitis (EP), 3) rats subjected to hyposalivation (H), and 4) rats concomitantly subjected to EP and H. Groups 2 and 4 received

20 µl injections of LPS (1 mg/ml) derived from *Escherichia coli* (serotype 055-B5, Sigma-Aldrich) into the vestibular and lingual gingiva of both the right and left first molars, and into the interdental space between the first and second molars (a total 60 µl of LPS per tooth, 120 µl per rat per treatment) while under sevoflurane inhalation anesthesia. This injection regimen, commonly used as an experimental periodontitis model, was carried out over six weeks, with injections administered on days 1, 3, and 5 of each week, following a previously established method. The gingival injections were delivered using a 13-mm 27-gauge microfine insulin syringe. Groups 3 and 4 underwent bilateral submandibulectomy surgery under anesthesia seven weeks before euthanasia, utilizing intraperitoneal injections of ketamine hydrochloride (Holliday-Scott SA, 70 mg/kg) and 2% xylazine hydrochloride (König Laboratories SA, 10 mg/kg), as described in previous studies. A 15-mm midline incision was made on the ventral neck, allowing the excretory ducts and major blood vessels to be ligated. The submandibular-sublingual salivary glands were carefully excised from surrounding connective tissue, preserving key neural structures, such as the marginal mandibular branch of the facial nerve, along with the hypoglossal and facial nerves. All experiments were repeated at least twice, with the graphs showing the outcomes of one of these trials.

### *Macroscopic assessment of periodontal bone loss: distance and width techniques*

Immediately following euthanasia, hemimandibles and hemimaxillae were dissected, cleaned, and stained with 1% aqueous methylene blue to highlight the cemento-enamel junction (CEJ) and the alveolar crest (AC)<sup>28</sup>. A stereomicroscope and a stainless-steel digital caliper were used to measure three buccal and three lingual/palatal distances (mesial, central and distal) between the CEJ and the AC. The sum of these three measurements on both sides of the upper and lower molars served as an indicator of alveolar bone loss (ABL), expressed in millimeters. The width of the mandibular alveolar process was determined at the level of the first mandibular molar. Using the digital caliper, the distance between two points located at the central root level on the buccal and lingual surfaces was measured in millimeters.

### Gene expression by semi-quantitative RT-PCR

For the gene expression analysis of CB1r and CB2r mRNA, tissue samples were obtained from the free and attached gingiva of the first molars and submandibular glands. For the gene expression analysis of RANKL and OPG mRNA, dental insertion tissues (including alveolar bone, periodontal ligament and root cementum) were collected by alveolar scraping performed after post-mortem extraction of the first molars. To ensure an adequate yield of total mRNA, tissue was harvested bilaterally from the maxilla and right hemimandible, and collected the equivalent of tissue from three teeth of the same animal. Total messenger RNA was isolated using RNeasy Reagent according to the manufacturer's protocol (Molecular Research Center Inc., Cincinnati, OH, USA). RNA concentration and purity were determined by measuring absorbance at 260 and 280 nm using a Pico200 Microliter UV/Vis spectrophotometer (Spectra Services Inc.). Subsequently, 2 µg of total RNA was reverse-transcribed into cDNA using the Improm-II™ system (Promega Corporation, Madison, USA) with Oligo(dT) primers (Invitrogen™) and a ribonuclease inhibitor (Promega, USA). Semi-quantitative PCR was carried out using GoTaq® DNA Polymerase (Promega, USA) on a GenePro thermal cycler (BIOER). The PCR protocol began with an initial denaturation at 94 °C for 2 minutes, followed by 25 to 30 cycles of 1 minute at 94 °C (denaturation), 1 minute at 60 °C (annealing), and 2 minutes at 72 °C (extension), with a final extension at 72 °C for 5 minutes. Primers specific to the target genes, listed in Table 1, were designed using the NCBI Primer-BLAST tool and synthesized by Biodynamics®. All gene expression results were normalized to β-actin, which was used as the reference gene. The PCR products were analyzed through electrophoresis on a 2% agarose gel, stained with GelRed™ nucleic acid stain (Biotium, Inc., Fremont, CA, USA), visualized with a Gel Doc XR+ imaging system (BioRad, CA,

USA), and quantified using Image Lab software. The data were presented in arbitrary units (AU) of relative optical density.

### Salivary secretion assessment

One week before euthanasia, rats from the control and EP groups were anesthetized via intraperitoneal injection of ketamine hydrochloride (Holliday-Scott SA, 70 mg/kg) and 2% xylazine hydrochloride (König Laboratories, 10 mg/kg). Then, pilocarpine (Sigma-Aldrich, 0.5 mg/kg) was administered intraperitoneally to induce salivation, and a cotton ball was promptly placed under the tongue to collect the entire salivary output. The amount of saliva was determined by weighing the cotton ball before and after the collection. The saliva was gathered over a 90-minute period after pilocarpine administration. The rats and the saliva were weighed, and salivary production was reported as milligrams of saliva secreted per gram of body weight.

### PGE<sub>2</sub> assessment by radioimmunoassay

To measure PGE<sub>2</sub> levels, submandibular gland samples were homogenized in 1 mL of absolute ethanol. After centrifugation, the supernatants were dried using a centrifugal vacuum concentrator (Speed Vac, Thermo Fisher Scientific) at room temperature. The resulting residues were resuspended in buffer, and antiserum (Sigma-Aldrich) was added according to the method outlined by Mohn et al., 2011<sup>29</sup>. The assay had a sensitivity of 12.5 pg per tube, with 100% cross-reactivity for PGE<sub>2</sub> and PGE<sub>1</sub>, and only 0.1% for other prostaglandins. The intra-assay and inter-assay variation coefficients for PGE<sub>2</sub> were 8.2% and 12.0%, respectively. The results were reported as picograms of PGE<sub>2</sub> per milligram of wet tissue weight.

### Statistical analysis

Data were presented as the mean of six determinations ± SEM for each group. Statistical

**Table 1. Primer sequence for specific markers.**

| GENE     | ACCESSION NUMBER | FORWARD PRIMER (5'-3') | REVERSE PRIMER (5'-3') |
|----------|------------------|------------------------|------------------------|
| β-ACTINA | NM_031144.3      | ACCCGCCGAGTACAACCTTC   | ATGCCGTGTTCAATGGGGTA   |
| RANKL    | NM_057149.1      | ACCAGCATCAAAATCCCAAG   | TTTGAAAGCCCCAAAGTACG   |
| OPG      | NM_012870.2      | GTTCTTGACAGCTTCACCA    | AAACAGCCCAGTGACCATTC   |
| CB1r     | NM_001429314.1   | AGGAGCAAGGACCTGAGACA   | TAACGGTGCTCTTGATGCAG   |
| CB2r     | NM_009924.4      | AGGTTGCATTCACCAACAGAC  | TTAGTTCCTCTGGGCAATGG   |

analysis was performed using Student's t-test for comparisons between two groups, and Two-Way ANOVA followed by Tukey's post-hoc test for unequal replicates. The sample size was selected to ensure an 80% power to detect differences between treatments comparable to the experimental error. All statistical tests were conducted using Prism software (GraphPad Software), with significance defined as P values less than 0.05.

## RESULTS

### Assessments in periodontal tissues

#### *Messenger RNA expression of CB1r and CB2r*

The expression of CB1r mRNA in gingival tissue homogenates was almost two times higher in groups EP, H and EP+H than in controls, while the expression of CB2r mRNA was more than three times higher in group EP and two and a half times higher in groups H and EP+H than in controls (Table 2). Additionally, CB2r mRNA expression was significantly higher in group EP than in group H.

**Table 2. Messenger RNA expression of CB receptors in gingival tissue measured by RT-PCR.**

| Group/<br>parameter | CB1r/ $\beta$ -actin<br>mRNA expression<br>(AU) | CB2r/ $\beta$ -actin<br>mRNA expression<br>(AU) |
|---------------------|-------------------------------------------------|-------------------------------------------------|
| Control             | 0.27 $\pm$ 0.06 <sup>A</sup>                    | 0.38 $\pm$ 0.04 <sup>A</sup>                    |
| EP                  | 0.46 $\pm$ 0.05 <sup>B</sup>                    | 1.22 $\pm$ 0.08 <sup>B</sup>                    |
| H                   | 0.51 $\pm$ 0.08 <sup>B</sup>                    | 0.98 $\pm$ 0.06 <sup>C</sup>                    |
| EP+H                | 0.49 $\pm$ 0.1 <sup>B</sup>                     | 1.041 $\pm$ 0.1 <sup>BC</sup>                   |

EP: experimental periodontitis. H: hyposalivation. Values are expressed as media  $\pm$  standard error. Different letters represent significant statistical differences analyzed by One Way ANOVA followed by Tukey's multiple comparisons test ( $p < 0.05$ ).

#### *Macroscopic assessment of periodontal bone loss*

##### *Distance and width methods*

Gingival administration of LPS (group EP) and SM (group H) increased alveolar bone loss evaluated by both distance and width methods. Bone loss determined as the increased distance between the CEJ and the AC was evidenced in groups EP, H and EP+H compared to controls, in both maxilla and mandible (Fig. 1A and Fig. 1B). Even in the mandible, bone loss was greater in group EP+H than in EP and H alone. However, the width method, which evaluates alveolar bone levels by measuring in the buccal-lingual direction on the mandibular first molars, confirmed that EP, H and EP+H conditions

induced bone loss compared to the control group. However, no significant differences were observed among the three experimental groups (Fig. 1C).

#### *Messenger RNA expression of RANKL and OPG*

RANKL mRNA expression in homogenized dental insertion tissues was significantly higher in groups EP and EP+H compared to the control group, while OPG mRNA expression was notably lower (Table 3). In contrast, group H did not differ significantly from the control group in RANKL or OPG mRNA expression. The RANKL/OPG ratio was similar between the H and control groups but was twice as high in groups EP and EP+H than in the other two groups.

### Assessments in submandibular glands

#### *Messenger RNA expression of CB1r and CB2r*

The expression of CB1r mRNA in SMG homogenates was almost two times higher in group EP than in controls (Table 4). It was noteworthy that even though the basal expression of CB2r mRNA was lower than that of CB1r, it was more than six times higher in group EP than in controls.

#### *Salivary secretion assessment*

After a 90-minute post-stimulation period, secreted saliva was significantly reduced in animals that received LPS gingival administration (group EP) than in controls (Fig. 2A).

#### *Radioimmunoassay of PGE<sub>2</sub>*

PGE<sub>2</sub> content level in submandibular glands, a known mediator of salivary flow reduction, was higher in the EP group than in controls (Fig. 2B).

## DISCUSSION

The relationship between oral diseases and cannabinoid receptors is an area of growing scientific interest. Previous studies by our group have demonstrated that the administration of cannabinoid receptor agonists exerts a therapeutic effect in experimental models of periodontitis, resulting in decreased alveolar bone loss and gingival inflammation<sup>24,30-32</sup>. *In vitro* studies using human periodontal cells further support these beneficial effects<sup>33</sup>.

In the present study, we investigated the expression levels of cannabinoid receptors in the periodontal tissues and salivary glands of rats subjected to

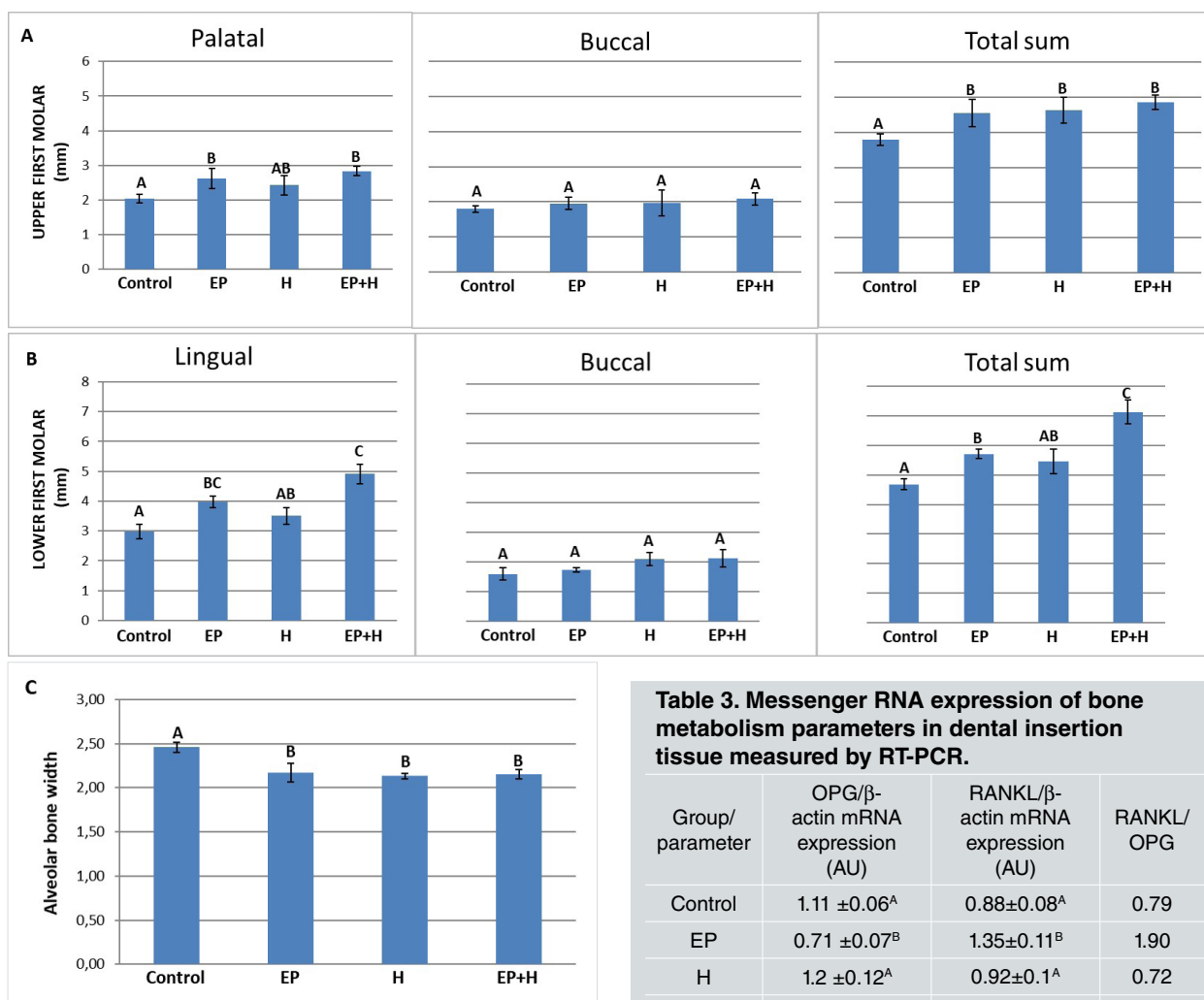


Fig. 1: Morphometric evaluation of alveolar bone loss in cortical plates: A) Palatal face, buccal face and total sum in the upper first molar. B) Lingual face, buccal face and total sum in the lower first molar. C) Width of the alveolar bone measured in the buccal-lingual direction in the lower first molars. EP: experimental periodontitis. H: hyposalivation. Results are presented as mean  $\pm$  SEM. Different letters represent significant statistical differences analyzed by One Way ANOVA followed by Tukey's multiple comparisons test ( $p < 0.05$ ).

experimental periodontitis and hyposalivation induced by submandibulectomy. Our results demonstrated upregulation of both CB1r and CB2r under these pathological conditions. These findings provide additional insight into the involvement of the endocannabinoid system (ECS) in oral disease pathogenesis and support its potential as a therapeutic target.

The upregulation of cannabinoid receptors in inflamed tissues, as shown in this study, may reflect a compensatory anti-inflammatory mechanism aimed at mitigating tissue damage during the inflammatory

**Table 3. Messenger RNA expression of bone metabolism parameters in dental insertion tissue measured by RT-PCR.**

| Group/parameter | OPG/ $\beta$ -actin mRNA expression (AU) | RANKL/ $\beta$ -actin mRNA expression (AU) | RANKL/OPG |
|-----------------|------------------------------------------|--------------------------------------------|-----------|
| Control         | 1.11 $\pm$ 0.06 <sup>A</sup>             | 0.88 $\pm$ 0.08 <sup>A</sup>               | 0.79      |
| EP              | 0.71 $\pm$ 0.07 <sup>B</sup>             | 1.35 $\pm$ 0.11 <sup>B</sup>               | 1.90      |
| H               | 1.2 $\pm$ 0.12 <sup>A</sup>              | 0.92 $\pm$ 0.1 <sup>A</sup>                | 0.72      |
| EP+H            | 0.87 $\pm$ 0.09 <sup>B</sup>             | 1.47 $\pm$ 0.15 <sup>B</sup>               | 1.68      |

EP: experimental periodontitis. H: hyposalivation. Values are expressed as mean  $\pm$  standard error. Different letters represent significant statistical differences analyzed by One Way ANOVA followed by Tukey's multiple comparisons test ( $p < 0.05$ ).

**Table 4. Messenger RNA expression of CB receptors in submandibular gland measured by RT-PCR.**

| Group/parameter | CB1r/ $\beta$ -actin mRNA expression (AU) | CB2r/ $\beta$ -actin mRNA expression (AU) |
|-----------------|-------------------------------------------|-------------------------------------------|
| Control         | 0.52 $\pm$ 0.09 <sup>A</sup>              | 0.23 $\pm$ 0.12 <sup>A</sup>              |
| EP              | 0.95 $\pm$ 0.13 <sup>B</sup>              | 1.48 $\pm$ 0.22 <sup>B</sup>              |

EP: experimental periodontitis. Values are expressed as mean  $\pm$  standard error. Different letters represent significant statistical differences analyzed by Student's T-test ( $p < 0.05$ ).

process. This interpretation aligns with previous findings in human tissues. For instance, Navarro-Saiz et al.<sup>34</sup> reported that both CB1r and CB2r are expressed in human odontoblasts and gingival fibroblasts, and that CB2r expression increases significantly under inflammatory conditions.

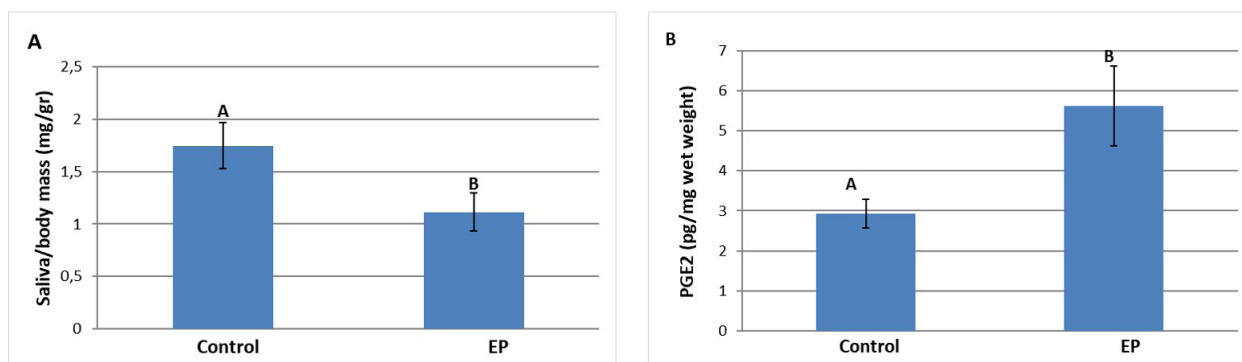


Fig. 2: A) Salivary secretion (mg of saliva/body mass) after administration of pilocarpine to rats in the control and EP groups. B) PGE<sub>2</sub> content (pg/mg wet weight) in the submandibular glands of rats from the control and EP groups. EP: experimental periodontitis. Values are expressed as mean  $\pm$  standard error. Different letters represent significant statistical differences analyzed by Student's *t*-test ( $p < 0.05$ ).

Similarly, Nakajima et al.<sup>3</sup> found higher CB2r expression in gingival fibroblasts from patients with gingivitis and periodontitis compared to healthy individuals<sup>3</sup>. In contrast, Ataei et al.<sup>35</sup> reported decreased cannabinoid receptor expression in periodontitis patients, suggesting that reduced ECS signaling might be linked to greater susceptibility to disease progression. However, this discrepancy could stem from different stages or mechanisms of disease and should be interpreted with caution.

In our study, rats with experimental periodontitis exhibited a significant increase in the RANKL/OPG ratio and notable alveolar bone loss, accompanied by elevated CB1r and CB2r mRNA levels in gingival tissues, particularly CB2r. These findings suggest that cannabinoid receptors may participate in endogenous homeostatic responses aimed at controlling inflammation and preventing further periodontal destruction. In the case of hyposalivation induced by submandibulectomy, CB1r and CB2r gingival expression was also increased, with concomitant alveolar bone loss, though without a significant change in the RANKL/OPG ratio. This suggests the activation of an alternative damage pathway, possibly indirect and different from the one triggered by LPS during periodontitis.

Although literature exploring cannabinoid receptor expression in inflamed oral tissues remains limited, evidence from other organ systems supports a similar modulatory role. For instance, CB2r upregulation has been observed in microglia following brain injury, contributing to neuroprotection by attenuating the inflammatory response<sup>36</sup>. Similarly, increased CB2r expression in synovial tissues after joint injury has been linked to anti-inflammatory effects

mediated through fibroblasts and macrophages<sup>37</sup>. These parallels reinforce the hypothesis that CB2r upregulation in oral tissues may play a protective, inflammation-limiting role.

With regard to the ECS in salivary glands, previous studies have shown that CB1r and CB2r are differentially expressed in murine submandibular glands<sup>14</sup>. CB1r was predominantly found in the ductal structures, while CB2r was located mainly around acinar cells. In that study, activation of cannabinoid receptors by anandamide was shown to inhibit stimulated salivary secretion, an effect reversed by selective antagonists. These findings suggest that the ECS exerts a regulatory influence on salivary gland function under physiological conditions. In this context, the reduced salivary response to pilocarpine observed in our experimental periodontitis model likely reflects a similar mechanism. This reduction, together with the six-fold increase in CB2r expression in salivary glands during periodontitis, strongly suggests that the ECS may be actively involved in the pathophysiological processes leading to decreased salivary secretion. Thus, the ECS could be contributing to the development of xerostomia frequently associated with periodontal disease, not only through modulation of secretion, but also by influencing local inflammatory responses. This highlights a potential modulatory role of the ECS in the context of chronic oral inflammation.

The physiological significance of cannabinoid receptor upregulation in oral tissues under inflammatory conditions may lie in the ECS's immunomodulatory and anti-inflammatory capabilities. Endogenous cannabinoids acting through CB1r and CB2r can suppress pro-

inflammatory mediator release and influence leukocyte activity (including that of neutrophils, lymphocytes and macrophages), thus regulating immune responses and preventing excessive tissue destruction<sup>4,5</sup>. Given that periodontitis is a chronic bacterial infection, increased receptor expression may serve as a protective mechanism to maintain tissue homeostasis and limit disease progression.

In summary, the observed upregulation of cannabinoid receptors in response to both periodontal inflammation and hyposalivation suggests a coordinated ECS-mediated response to pathophysiological stress. In periodontal tissues, this may be an attempt to reduce inflammation and preserve tissue integrity. In salivary glands, while the situation is more complex, the ECS may contribute to both immune regulation and salivary secretion. Prior findings support the hypothesis that cannabinoid-mediated inhibition of salivary

secretion in inflamed conditions could be a defense mechanism to minimize the dissemination of pathogens and inflammatory agents, particularly via CB1r<sup>23</sup>. Conversely, CB2r appears to mediate the anti-inflammatory and modulatory responses most prominently observed under our experimental conditions.

Cannabinoid receptor upregulation during chronic oral inflammation likely constitutes a homeostatic feedback mechanism aimed at restoring ECS balance. The present and prior findings of our group support the potential development of cannabinoid-based therapies to reduce inflammation, protect periodontal tissues and facilitate tissue repair. As research in this field is still emerging, additional studies are warranted to elucidate further the regulatory dynamics of cannabinoid receptors in oral pathologies and to explore their therapeutic potential.

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## CONFLICT OF INTERESTS

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

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# Assessment of two designs for an Oral surgery Postoperative Leaflet

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## ABSTRACT

Around the world it is considered good practice to provide patients with a postoperative instruction and advice leaflet following tooth extractions. It has never been reported whether the design or the content is effective or helpful for a patient's recovery. **Aim:** To assess the opinions of patients about two postoperative instructions leaflets with similar content but different designs. **Materials and Method:** A questionnaire was administered to 32 patients who had recently undergone oral surgery. The questionnaire asked patients about their satisfaction with the information and their overall opinion of two postoperative leaflet designs (a conventional and a daily planner design). **Results:** 53% of the patients preferred the daily planner design and 6.3% preferred the conventional model. 60% of the patients recommended the daily-planner leaflet for future patients. **Conclusion:** The results of this study suggest that patients find postoperative instructions leaflets to be helpful because they can help to guide patient to follow instructions after surgery. The daily planner was preferred because it was easy to review every day during recovery.

**Keywords:** oral surgery - postoperative instructions - leaflet - procedures

## Estudio comparativo aleatorizado de dos diseños de folletos postoperatorios de cirugía oral

## RESUMEN

Alrededor del mundo es una buena práctica proporcionar a los pacientes un panfleto o folleto con instrucciones y consejos postoperatorios después de una cirugía bucal. Hasta la fecha no hay reporte sobre si el diseño o el contenido es efectivo o ayudó a los pacientes en su recuperación. **Objetivo:** Evaluar las opiniones de los pacientes sobre dos folletos de instrucciones postoperatorias con contenido similar pero diferentes diseños. **Materiales y Método:** Se administró un cuestionario a 32 pacientes que se habían sometido recientemente a cirugía bucal. El cuestionario preguntó a los pacientes sobre su satisfacción con la información y su opinión general de cada uno de los dos diseños de folletos postoperatorios (un diseño convencional y un diseño similar a una agenda o calendario). **Resultados:** El 53% de los pacientes prefirió el diseño de agenda/calendario y el 6,3% el modelo convencional. El 60% de los pacientes recomienda usar el folleto de agenda/calendario con futuros pacientes. **Conclusión:** Los resultados de este estudio sugieren que los pacientes encuentran útiles los folletos de instrucciones postoperatorias. Ya que pueden ayudar a guiar y seguir las instrucciones después de la cirugía. El calendario/agenda fue el preferido por ellos, ya que les resultó fácil revisarlo todos los días durante la recuperación.

**Palabras clave:** cirugía oral - postoperatorio - instrucciones - prospecto - procedimientos

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## INTRODUCTION

The second principle in the latest review of the British General Dental Council's (GDC) 9 principles for standard practice in the United Kingdom states that dentists must communicate effectively with their patients, specifying *"You need to ensure that the patients have understood the information you have given them. After minor oral surgery (MOS), it is good practice to provide patients with a leaflet/pamphlet with instructions on how to care for their mouth during recovery"*<sup>1</sup>.

Studies cited by Kessels and Alvira-Gonzalez report that 40 to 80% of medical information provided by healthcare practitioners is forgotten immediately. This could be the result of several factors including stress, level of education, age and the perceived importance or focus of the information provided<sup>2,3</sup>.

Most patients prefer their own doctors to explain instructions rather than watching a pre-recorded video<sup>2</sup>. This explains why audiovisual alternatives such as pre-recorded videos have mixed results in patient compliance<sup>2-4</sup>. Communication should be consistent and effective, recognising patients' communication difficulties, using direct, specific instructions, graphics and pictographics, and avoiding professional jargon and acronyms<sup>1-4</sup>.

Most oral surgery postoperative leaflets follow the same conventional designs which have been used from the 1940s or even as early as the 1920s in the various options produced by modern institutions and science college<sup>5-8</sup>. This means that for the last 80 years, the same leaflet design has been used over and over<sup>5-8</sup>.

Motivated by the desire to increase patient compliance, reduce the risk of postoperative complications, and limit the number of patients phoning our hospital to seek clarification of or advice on the postoperative instructions, I have proposed a new leaflet based on other studies and psychological experiences.

## MATERIALS AND METHOD

This was a randomised comparative study on 50 patients who had recently undergone MOS. Based on their availability, patients were randomly assigned by the booking team to a specific MOS session that would provide two post-operative instructions leaflets: a conventional leaflet and a newly designed calendar/daily planner leaflet. This

was done independently of the patient's gender, age, type of procedure, type of anaesthetic (intravenous sedation or local anaesthetic) and medical history. Neither the booking team nor the staff in the surgical theatre were aware that the postoperative package included 2 different types of leaflets.

The conventional leaflet (Fig. 1) is a double-sided A4 sheet with portrait orientation containing information divided into sentences with specific instructions related to pain, swelling, bleeding, oral hygiene, diet, stitches, medication, rest, follow-up, contact instructions and a frequently asked question section for managing complications.

The calendar/daily-planner leaflet is a landscape-oriented A4 sheet folded in half like a booklet, providing four pages of content. The information is similar to that in the previous leaflet but distributed in the form of a calendar or daily planner, where instead of being divided into paragraphs, the instructions are divided by days as "dos and don'ts" during recovery, along with tips to keep in mind during each specific day or any complication that may arise (Fig. 2). The back of the leaflet shares space with the cover and provides further advice on how to deal with pain, and information on the relevance of early chewing function in relation to quality of life (Fig. 3). The models shown in Figures 1, 2 and 3 are coloured but the versions provided to the patients were in shades of grey.

Once the new leaflet model and questionnaire were approved by all Consultants and Nursing staff involved in the care of MOS patients, it was distributed to the patients along with the conventional leaflet model. Patients received both leaflets at the same time.

Two to three weeks later, a questionnaire was sent electronically to these patients so they could provide their opinion about each leaflet. The time between phases was to allow the patients to fully recover from their surgeries. The final intention of the questionnaire was to assess which leaflet made more impact and was more helpful during recovery. We did not assess information retention or compliance. A binomial test / chi square goodness of fit was used to test whether there was significant preference for one of the leaflets. Even with a small sample, the difference was statistically significant due to the disproportionate preference for one of the leaflets.



**Practice Plus Group**



Practice Plus Group Hospital, Southampton

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**FREQUENTLY ASKED QUESTIONS FOLLOWING DENTAL SURGERY**

- I have swelling. Is this normal?**  
Yes, moderate swelling could occur 2-3 days after your surgery, place a cold compress against the area a few times a day to help, any excessive swelling or swelling of your neck may be a concern, please call us
- I have noticed there is a hole where the tooth was and I did not have stitches. Is this normal?**  
Yes, it is not always necessary to have stitches; the consultant on the day of surgery would make the decision as to whether or not you needed stitches.
- The area is painful. Is this normal?**  
Yes. The area should be painful for up to two weeks after the extraction. Often the pain could set in or get a little worse on day 3-4 after your surgery, please refer to your post-op instructions and manage this with pain relief  
If we prescribed painkillers for you to take home, please make sure you are taking them regularly. If we gave you antibiotics it is vital that you finish your course to prevent further problems.
- The area is bleeding. Is this normal?**  
Minimal bleeding especially after eating and drinking is normal, please control this with the gauze you were given when discharged. If there is excessive bleeding or you are taking medication to thin your blood, please call us if you are concerned.

If you do feel the need to see a consultant, please ensure you call us first (phone number below), please do not just turn up as this will cause delays to other patients and you may not be seen.

**Additional information**

---

For the first 6 weeks after your procedure we will be happy to assist you should you have any problems with your mouth.

**Monday – Friday 8am – 5:00pm call 023 8071 2028 – please leave a message, you will be called back.**

**After 5pm and weekends for advice call 0333 999 2564 – PLEASE NOTE NO DENTIST ON SITE AT THIS TIME.**

**ORAL SURGERY DEPARTMENT**

**CARE OF THE MOUTH AFTER MINOR ORAL SURGERY PROCEDURE**

**Local anaesthetic**

The local anaesthetic used during the procedure may take a few hours to wear off. The anaesthetic will affect the gum close to the tooth but may also cause numbness of the lip, chin or tongue whilst they are numb.

You may eat as normal but be careful that food and drink are no too hot that they burn the numb areas, or disturb the surgical area.

Avoid sharp or crunchy foods (ie toast, crisps) for 2-3 days as these will also disturb healing and may cause bleeding.

**BLEEDING**

It is normal for there to be some oozing from the socket/surgical area after the procedure. Your saliva may be slightly bloodstained for a few days. To prevent further bleeding you should return home after the procedure and rest, if the bleeding persists, apply pressure for 20-30 minutes on the area either using the pack you have been given or a clean handkerchief slightly dampen this before use.

**PAIN RELIEF**

We recommend that you take painkillers for the first few days after the procedure it is normal to require painkillers for up to two weeks after surgery, you should also expect some swelling around the area following surgery, if you are not prescribed painkillers by us, Paracetamol or Ibuprofen based preparations should be used.

**FOLLOWING THE PROCEDURE**

**TODAY** – Do not rinse your mouth or spit out as this can disturb the healing socket. Tooth brushing should be carried out as normal, but extra care should be taken around the extraction site.

**TOMORROW** – Dissolve a teaspoon of salt in a little warm water and then begin bathing the area, hold the solution over the area for a minute. This should be carried out as often as possible, or at least after every meal and continued for 10 days. Corsodyl mouthwash can be used as an alternative.

The tooth socket may feel uneven or lumpy for the first few months, this is entirely normal.

**SMOKING AND ALCOHOL**

Smoking greatly increases the risk of infection and slows the healing process it also increases the chances of localised bone infection, this is an extremely painful condition which can prove difficult to treat. DO NOT SMOKE UNTIL THE WOUND HAS FULLY HEALED

**ALCOHOL SHOULD BE AVOIDED FOR 24 HOURS AS IT MAY INCREASE BLEEDING**

**STITCHES** can take up to three weeks to dissolve, if any stitches come out before dissolving please do not be concerned unless the area begins to bleed.

Fig. 1: Front and back content of the conventional leaflet.



## Your Post Operation calendar

Tick the box on each day task when done.

| 1st Day                                                                                                          |                                                                                         | 2nd Day                                                                                                            |                                                                        | 3rd Day                                                                                                       |                                                                                           | 4th & 5th Day                                                                       |                                                                                     | 7th Day                                                   |                                                                                   | 10th Day                                                                                |                                                                                                                                |                                                                                                                                         |                                                  |
|------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------|-----------------------------------------------------------|-----------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------|
| DO                                                                                                               |                                                                                         | DO                                                                                                                 |                                                                        | DO                                                                                                            |                                                                                           | DO                                                                                  |                                                                                     | DO                                                        |                                                                                   | DO                                                                                      |                                                                                                                                |                                                                                                                                         |                                                  |
| <input type="checkbox"/> Brush your teeth. Not the wound or the stitches.                                        | <input type="checkbox"/> Take pain relief as advised by your nurse/Dr (Soft or liquid). | <input type="checkbox"/> Have something to eat (Soft or liquid).                                                   | <input type="checkbox"/> Use cold packs over the skin.                 | <input type="checkbox"/> Continue Oral hygiene and on this day you can also rinse your mouth and spit gently. | <input type="checkbox"/> Continue pain relief.                                            | <input type="checkbox"/> START to eat soft and semisolid food as tolerated.         | <input type="checkbox"/> Continue brushing and rinsing your mouth after every meal. | <input type="checkbox"/> Continue pain relief.            | <input type="checkbox"/> Continue to eat semi-solid food as tolerated.            | <input type="checkbox"/> Use Warm packs over the skin.                                  | <input type="checkbox"/> Continue brushing and rinsing your mouth after every meal.                                            | <input type="checkbox"/> Start a normal diet including hard food if tolerated.                                                          | <input type="checkbox"/> Can start light sports. |
| <b>DON'T</b>                                                                                                     |                                                                                         | <b>DON'T</b>                                                                                                       |                                                                        | <b>DON'T</b>                                                                                                  |                                                                                           | <b>DON'T</b>                                                                        |                                                                                     | <b>DON'T</b>                                              |                                                                                   | <b>DON'T</b>                                                                            |                                                                                                                                |                                                                                                                                         |                                                  |
| <input type="checkbox"/> Don't spit or rinse your mouth.                                                         | <input type="checkbox"/> Don't smoke.                                                   | <input type="checkbox"/> Don't do any sports activities, hot food, drinks or hot showers.                          | <input type="checkbox"/> Don't use straws.                             | <input type="checkbox"/> Don't smoke.                                                                         | <input type="checkbox"/> Don't do any sports activities, hot food, drinks or hot showers. | <input type="checkbox"/> After 24 hours don't use cold packs over the skin anymore. | <input type="checkbox"/> Don't smoke.                                               | <input type="checkbox"/> Don't do any sports.             | <input type="checkbox"/> Don't smoke.                                             | <input type="checkbox"/> Don't take any pain relief tablets unless you are having pain. |                                                                                                                                |                                                                                                                                         |                                                  |
| <b>REMEMBER</b>                                                                                                  |                                                                                         | <b>REMEMBER</b>                                                                                                    |                                                                        | <b>REMEMBER</b>                                                                                               |                                                                                           | <b>REMEMBER</b>                                                                     |                                                                                     | <b>REMEMBER</b>                                           |                                                                                   | <b>REMEMBER</b>                                                                         |                                                                                                                                |                                                                                                                                         |                                                  |
| <input type="checkbox"/> It will be sore and painful, specially while eating and brushing but you need to do it. | <input type="checkbox"/> You will have taste of blood.                                  | <input type="checkbox"/> Today its going to be more swollen, painful and more uncomfortable. Bruises might appear. | <input type="checkbox"/> This should be the last day with blood taste. | <input type="checkbox"/> Today the swelling and pain should reach its peak point.                             | <input type="checkbox"/> Still very uncomfortable. Be patient. Allow it to heal properly. |                                                                                     |                                                                                     | <input type="checkbox"/> Swelling should start to reduce. | <input type="checkbox"/> Do jaw exercises (open and close to prevent joint pain). | <input type="checkbox"/> Swelling should be gone by now.                                | <input type="checkbox"/> Do jaw exercises (open and close to prevent joint pain). No need to call unless it is too unbearable. | <input type="checkbox"/> The tooth socket may feel uneven for the first few months, Stitches can take 3-4 weeks to dissolve completely. |                                                  |

Fig. 2: Daily planner / Calendar leaflet content and design.

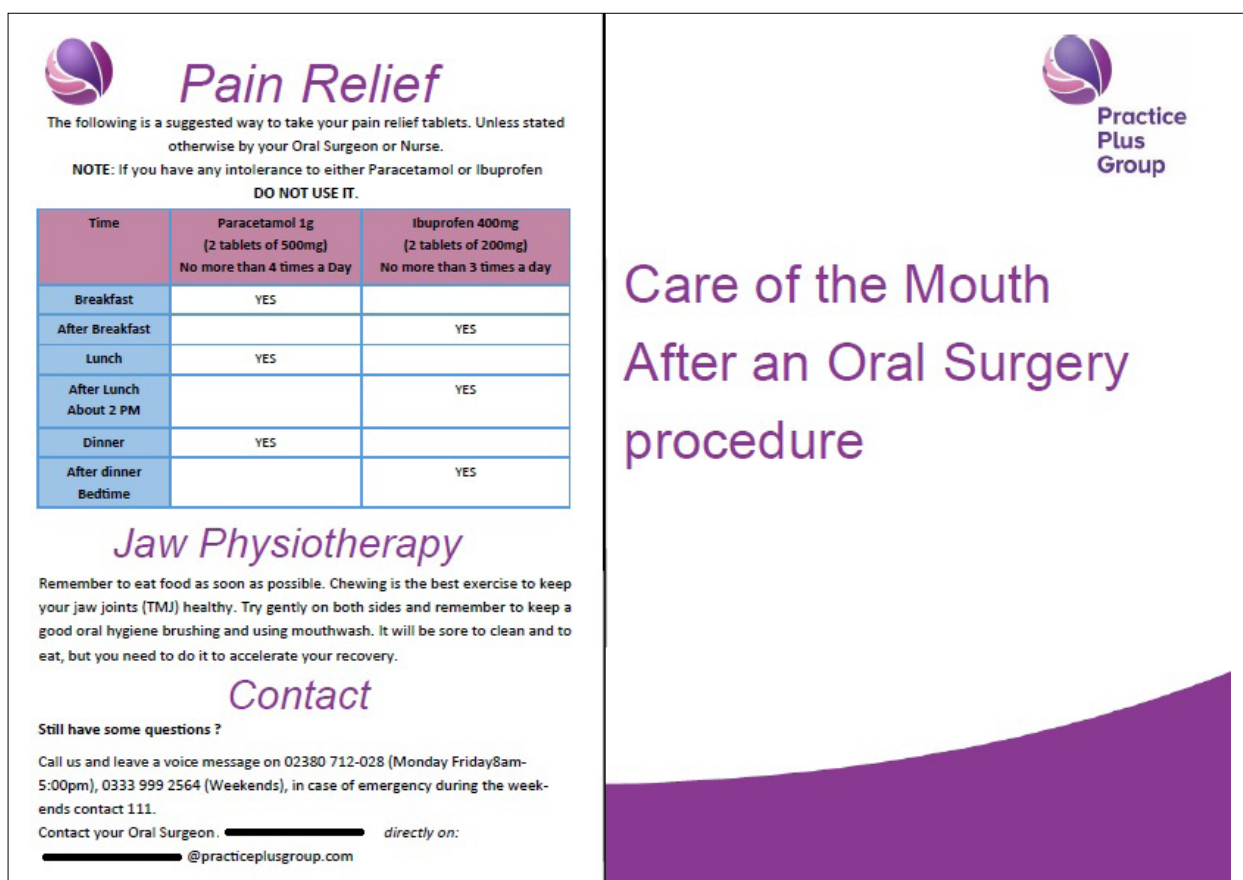


Fig. 3: Front cover and back page of the calendar leaflet.

## RESULTS

Out of 50 patients who were invited to complete the questionnaire, 33 responded. The data were entered in a Microsoft Excel spreadsheet for analysis. When asked which leaflet they found more helpful for their recovery, 54.5% (n=18) selected the calendar/daily planner leaflet, while only 6.0% (n=2) preferred the conventional leaflet. The remaining responses included 24.2% (n=8) who reported they did not read either leaflet and 15.2% (n=5) who stated both leaflets were the same (Table 1).

To assess whether the calendar leaflet was significantly preferred to the conventional design, a chi-square goodness-of-fit test was performed on the 20 patients who expressed a clear preference for either format. The test yielded  $\chi^2 = 12.80$ ,  $p < 0.001$ , indicating a statistically significant preference for the calendar.

The second question of the survey was “Did you find the answer to every problem you had during recovery in the leaflets? And in which one?”. The calendar/daily planner was the preferred answer with 42.4% (n=14), followed by “yes” on both with

24.2% (n=8), while 18.2% (n=6) felt that they did not have any problems during recovery, 9.1% (n=3) were unable to find the answer to their recovery problem in either leaflet, and only 6% (n=2) had their recovery issue resolved by the conventional leaflet only.

The following question was “Do you think the leaflet provides enough guidance to prevent you from calling our staff to ask a question? Which one was helpful for this?”. Most of the patients reported that they did not have any problem during recovery 37.5% (n=12). Out of those who had some issues during their postoperative period, 34.4% (n=11) indicated the calendar/daily planner was their main source of answers during recovery, and only one patient (3.1%) said that they used the conventional leaflet for this. Finally, 15.6% (n=5) of the respondents said they were unable to find their answer and still had to contact the hospital for additional help. The remaining 9.3% (n=4) provided various or incomplete responses.

**Table 1. Response summary of the questionnaire provided to the patients.**

| Questions                                                                                                                                                                                                                                                | Answers                                                                                      |                                                          |                                                               |                                           |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------|----------------------------------------------------------|---------------------------------------------------------------|-------------------------------------------|
| <b>1) After your surgery, you were given 2 post op instruction leaflets. One was a long set of instructions with a Frequently Asked Questions section, and the other one was like a calendar. Which one did you find more helpful for your recovery?</b> | The Calendar/day-planner model<br>54.5%<br>(n=18)                                            | Neither<br>39.4%<br>(n=13)                               | Conventional model<br>6.1%<br>(n=2)                           |                                           |
| <b>2) Did you find the answer to every problem you had during recovery on the leaflets? And in which one?</b>                                                                                                                                            | Yes, in the calendar leaflet<br>42.4% (N=14)<br>Yes, in the Conventional model<br>6.1% (n=2) | Yes, in both<br>24.2% (n=8)                              | No / Didn't have any problem<br>18.2% (N=6)                   | No, couldn't find my answer<br>9.1% (n=3) |
| <b>3) Do you think the leaflet guided you enough to prevent you from calling to ask our staff a question? Which one was helpful for this?</b>                                                                                                            | No, because I didn't have any problems during recovery<br>37.5%<br>(n=12)                    | Yes, the calendar was helpful enough.<br>34.4%<br>(n=11) | Neither, I still needed to call for advice.<br>15.6%<br>(n=5) | Various answers<br>12.5%<br>(n=5)         |
| <b>4) Between the 2 post op instruction leaflets, which one would you recommend we use for future patients?</b>                                                                                                                                          | Calendar leaflet<br>54.5%<br>(n=18)                                                          | Neither<br>21.2%<br>(n=7)                                | Both<br>18.2%<br>(n=6)                                        | Conventional model<br>6.1%<br>(n=2)       |

Finally, the patients were asked to recommend one of the two postoperative instruction leaflets for us to use with future patients. In reply, 54.5% (n=18) recommended the calendar/daily-planner, while only 6.1% (n=2) recommended the conventional design. Other answers included Neither 21.2% (n=7) and Both 18.2% (n=6).

Space was provided after every question for patients to write comments related to each answer. Twenty-seven patients thought a leaflet helped them to retain and remember all the postoperative information provided and was helpful during recovery.

When asked why they chose the calendar/daily planner over the conventional leaflet, some of the comments were:

*"Better layout"*

*"It was good to guide me by the day".*

*"Broken down, the information easier".*

*"Easy to follow".*

*"While I was recovering, I was able to look at the timetable, and it told me what to expect".*

*"The other one had more information, but the timetable was an easy way to remind me of things".*

*"It was easier to know what to do by the day".*

## DISCUSSION

A textbook for students on extraction of teeth written by S.S. Hornor in 1851 recommends giving postoperative instructions following an oral surgery

procedure, showing how important they are and how long they have been used. Although the book does not advocate the use of written information, it highlights the instructions and how the dentist should monitor the patient closely for the next 48 hours to ensure bleeding stops and pain is not increasing<sup>7</sup>.

The earliest leaflet I found during this investigation was in the American Dental Association (ADA) Library & Archives. It is a 4-page booklet published in 1920 and distributed by the "Library Bureau" (former name of the ADA library), called *"Postoperative Instructions to Dental Patients"*<sup>5</sup>. It is divided into 4 sections that explain the importance of rest, oral hygiene, diet, whether to use heat or cold and what to do in case of emergency. It also requests patients to contact the dentist's office on the first or second postoperative day to report progress of recovery<sup>5</sup>.

The next earliest pamphlet in the ADA Archives on this topic is called *"What to Do After Extraction of a Tooth"*, dated 1957, and produced and distributed by the ADA Bureau of Dental Health Education. It is also a 4-page booklet that divides the information into 5 sections: rest, cold applications, bleeding, oral hygiene and diet<sup>8</sup>.

Current leaflets are structured similarly to those from the olden days. Most leaflets available online from dental societies or professional organisations such as the British Association of Oral Surgeons (BAOS)



## Your Post Operation calendar

Tick the box on each day task when done

| 1st Day                                                                                                                                                                                                                                                           | 2nd Day                                                                                                                                                                                                                                        | 3rd Day                                                                                                                                                                                                                 | 4th & 5th Day                                                                                                                                                                                                                                                   | 6th to 9th Day                                                                                                                                                                                                            | 10th Day                                                                                                                                                                                                                       |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>DO</b>                                                                                                                                                                                                                                                         | <b>DO</b>                                                                                                                                                                                                                                      | <b>DO</b>                                                                                                                                                                                                               | <b>DO</b>                                                                                                                                                                                                                                                       | <b>DO</b>                                                                                                                                                                                                                 | <b>DO</b>                                                                                                                                                                                                                      |
| <input type="checkbox"/> Brush your teeth. Not the wound or the stitches.<br><input type="checkbox"/> Take pain relief.<br><input type="checkbox"/> Have something to eat (Soft or liquid).<br><input type="checkbox"/> Use cold packs over the skin.             | <input type="checkbox"/> Continue Oral hygiene and on this day you can also rinse your mouth and spit gently.<br><input type="checkbox"/> Continue pain relief.<br><input type="checkbox"/> START to eat soft and semisolid food as tolerated. | <input type="checkbox"/> Continue brushing and rinsing your mouth after every meal.<br><input type="checkbox"/> Continue pain relief.<br><input type="checkbox"/> Continue to eat soft and semisolid food as tolerated. | <input type="checkbox"/> Continue brushing and rinsing your mouth after every meal.<br><input type="checkbox"/> Continue pain relief.<br><input type="checkbox"/> Continue to eat semi-solid food as tolerated.<br><input type="checkbox"/> Use Warm packs over | <input type="checkbox"/> Continue brushing and rinsing your mouth after every meal.<br><input type="checkbox"/> Start a normal diet including hard food if tolerated.<br><input type="checkbox"/> Can start light sports. | <input type="checkbox"/> You should be feeling back to normal. That mean your recovery is complete.<br><br><b>Congratulations !!</b>                                                                                           |
| <b>DON'T</b>                                                                                                                                                                                                                                                      | <b>DON'T</b>                                                                                                                                                                                                                                   | <b>DON'T</b>                                                                                                                                                                                                            | <b>DON'T</b>                                                                                                                                                                                                                                                    | <b>DON'T</b>                                                                                                                                                                                                              | <b>REMEMBER</b>                                                                                                                                                                                                                |
| <input type="checkbox"/> Don't spit or rinse your mouth.<br><input type="checkbox"/> Don't smoke.<br><input type="checkbox"/> Don't do any sports activities, hot food, drinks or hot showers.<br><input type="checkbox"/> Don't use straws.                      | <input type="checkbox"/> Don't smoke.<br><input type="checkbox"/> Don't do any sports activities, hot food, drinks or hot showers.<br><input type="checkbox"/> After 24hours don't use cold packs over the skin anymore.                       | <input type="checkbox"/> Don't smoke.<br><input type="checkbox"/> Don't do any sports activities, hot food, drinks or hot showers.                                                                                      | <input type="checkbox"/> Don't smoke.<br><input type="checkbox"/> Don't do any sports.                                                                                                                                                                          | <input type="checkbox"/> Don't smoke.<br><input type="checkbox"/> Don't take any pain relief tablets unless you are having pain.                                                                                          | <input type="checkbox"/> The tooth socket may feel uneven or sharp for the first few months. this is the bone that was around the tooth, allow more time for this bone to smooth on its own (This can take at least 3 months). |
| <b>REMEMBER</b>                                                                                                                                                                                                                                                   | <b>REMEMBER</b>                                                                                                                                                                                                                                | <b>REMEMBER</b>                                                                                                                                                                                                         | <b>REMEMBER</b>                                                                                                                                                                                                                                                 | <b>REMEMBER</b>                                                                                                                                                                                                           |                                                                                                                                                                                                                                |
| <input type="checkbox"/> It will be sore and painful, specially while eating and brushing but you need to do it.<br><input type="checkbox"/> You will have taste of blood. In case of bleeding, apply pressure for 20 -30minutes on the area by biting the gauze. | <input type="checkbox"/> Today its going to be more swollen, painful and more uncomfortable. Bruises might appear.<br><input type="checkbox"/> This should be the last day with blood taste.                                                   | <input type="checkbox"/> Today the swelling and pain should reach its peak point.<br><input type="checkbox"/> Still very uncomfortable. Be patient. Allow it to heal properly.                                          | <input type="checkbox"/> Swelling should start to reduce.<br><input type="checkbox"/> Do jaw exercises (open and close to prevent joint pain).                                                                                                                  | <input type="checkbox"/> Swelling should be gone by now.<br><input type="checkbox"/> Do jaw exercises (open and close to prevent joint pain). No need to call unless it is too unbearable.                                | <input type="checkbox"/> Stitches can take 3-4 weeks to dissolve completely.<br><input type="checkbox"/> Scar tissue in the mouth looks white. This wont give you bad taste or bad smell.                                      |

Fig. 4: Daily planner / Calendar leaflet added content after this investigation.

are similar, designed either as a 4-page booklet with 1- or 2-sided pages containing specific instructions and different items related to main topics as pain, bleeding, smoking, alcohol, emergency care, rest and oral hygiene<sup>6</sup>. The conventional leaflet used in this study has a similar structure.

Kim et al. (2020) assessed the quality of information in modern postoperative leaflets providing instructions for patients following minor oral surgery procedures. They concluded that most of them contained low-quality information lacking details of any risks or consequences if the instructions were not followed, and the impact of overall quality of life during recovery. Most of them do not include dates or cite the articles or sources that support their content. This is no surprise, as the text and the idea are almost 100 years old. Kim et al. suggest reformatting and improving the quality of the information provided to patients<sup>9</sup>.

The new calendar/daily-planner was designed considering smart use of space, a single sheet of paper, direct simple instructions, and a different layout that would easily guide the patient through recovery<sup>6,7,9</sup>.

With the purpose of complementing the information provided verbally, our leaflet follows design advice from authors such as Kessels, Akshaya and the GDC Guidelines, in direct, easy to understand language. Moreover, the calendar-style layout graphically guides patients through recovery. We did not include bibliographical references because we could not afford to sacrifice valuable space that could be used to provide more information to patients<sup>6,7,9</sup>.

A non-published internal survey in our department confirmed that the most common reason for which patients contact our department is pain, followed by trismus and infections, with bleeding coming last. Based on these results, the calendar/daily planner design includes the usual advice following oral surgery but also emphasises pain management, oral hygiene and early chewing function to ensure faster recovery, a principle applied in AOCMF maxillofacial trauma<sup>10</sup>.

Ravindra et al. (2012) highlight how efficacy within the British National Health Service is often achieved with simple, practical improvement to clinical practice, such as updating a preoperative leaflet into

simpler, more direct design and language, ensuring preoperative patient compliance for sedation<sup>11</sup>. Our own experience has confirmed that at least 34.4% of the patients that had problems during recovery were able to find a way to solve them, while 3.1% of the patients reported the same for the conventional design. Based on the free text feedback from patients and the phone call made after implementation of these new leaflets, a new daily planner was prepared

containing additional information (Fig. 4).

## CONCLUSION

The results of our study suggest that in practice, patients prefer shorter, more concise, more basic language leaflets that provide easy reading and simple instructions to follow each day during their recovery, reducing the chances of needing to contact the Hospital.

## ACKNOWLEDGMENTS

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## CONFLICT OF INTERESTS

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

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# Influence of Bio-C and AH Plus sealers on bond strength determined by push-out of a fiberglass post installed with self-adhesive sealer

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## ABSTRACT

Given the limitations of the intraradicular adhesion procedure and the significant challenge of achieving effective bonding, it is essential to assess the potential impact of various endodontic sealers on the bond strength between fiberglass posts and root dentin. **Aim:** The aim of this study was to evaluate the bond strength of fiberglass posts to the root dentin of teeth filled with different endodontic sealers. **Material and Methods:** Forty-five single-rooted teeth were instrumented, filled using the single-cone technique, and divided into three groups according to the selected sealer (n=15): Bio-C sealer (GBC), AH Plus (GAH), and control group without sealer (GC). The post was installed using RelyX U200 self-adhesive resin sealer. Specimens were cut into a cervical, middle and apical thirds, to analyze one slice per third. Bond strength was evaluated using a push-out extrusion test. The failure modes in each third were observed under the microscope, and classified as adhesive, cohesive or mixed. **Results:** It was found a difference between groups in the cervical and apical thirds, with GAH having stronger adhesion in the cervical third than GBC (p=0.0015), although there was no difference with GC. In the apical third, GAH had higher adhesion values than GBC (p=0.0014) and GC (p=0.0005). In the middle third, there was no difference between groups (p=0.1386). The same sealer in the different thirds only differed significantly in the GBC group, where bond strength was higher in the middle and apical thirds, with a significant difference from the cervical third (p<0.05). There was no difference between failure types in any of the groups (p>0.05). **Conclusion:** The predominant failure between dentin and sealer was adhesive. It was concluded that the use of AH Plus endodontic sealer resulted in less impairment of the bond strength of resin cemented fiberglass posts compared to bioceramic sealer.

**Keywords:** dentin sealers - endodontics - root canal filling materials - resin cements - post and core technique

## Influência dos selantes Bio-C e AH Plus na resistência de união de rententores de fibra de vidro através do teste de push-out usando um selante autoadesivo

### RESUMO

Dadas as limitações do procedimento de adesão intraradicular e o significativo desafio de alcançar uma adesão eficaz, é essencial avaliar o impacto potencial de diversos cimentos endodônticos na resistência de união entre pinos de fibra de vidro e a dentina radicular. **Objetivo:** O objetivo deste estudo foi avaliar a resistência de união do pino de fibra de vidro à dentina radicular em dentes obturados com dois cimentos endodônticos, por meio do teste push-out. **Material e Métodos:** Foram instrumentados 45 dentes unirradiculares, obturados pela técnica do cone único, divididos em três grupos de acordo com o cimento selecionado (n=15), Bio-C Sealer (GBC), AH Plus (GAH) e o grupo controle, sem cimento (GC). O espaço do canal radicular cimentado com o pino+ cimento resinoso autoadesivo RelyX U200, seccionados, uma fatia em cada terço, em terços cervical, médio e apical. A resistência de união foi avaliada por meio do teste por extrusão push-out e os modos de falha observados em microscópio, sendo classificados em adesiva, coesiva e mista nos terços cervical, médio e apical. **Resultados:** Os resultados mostraram diferença entre os grupos no terço cervical e apical, no qual o GAH teve maior adesão que o GBC no terço cervical (p=0.0015), sem diferença do GC. E no terço apical o GAH teve maiores valores de adesão que o GBC (p=0.0014) e GC (p=0.0005). No terço médio não houve diferença entre os grupos (p=0.1386). Comparando o mesmo cimento em terços diferentes, houve diferença significativa somente no grupo GBC que apresentou maior resistência de união no terço médio e apical com diferença significativa em relação ao terço cervical (p<0.05). Não houve diferença entre os tipos de falhas em todos os grupos avaliados (p>0.05) e foi observado um predomínio de falha adesiva entre dentina e cimento. **Conclusão:** Concluiu-se que, o uso do cimento endodôntico AH Plus propiciou menos interferência na resistência de união de pinos de fibra de vidro cimentados com o cimento resinoso quando comparado ao cimento biocerâmico.

**Palavras-chave:** adesivos dentários - endodontia - materiais para obturação de canal radicular - cimentos resinosos - pinos de retenção dentária

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## INTRODUCTION

The clinical and biological success of endodontic treatment depends on a sequence of procedures, including root canal disinfection and obturation, sealing access to the cavity, and amount of remaining dentin<sup>1</sup>. Endodontic sealer is used for root canal obturation to promote the interface between dentin and gutta-percha<sup>2</sup>. The obturation phase is challenging due to the difficult adhesion between the proposed obturation materials and the walls of the root canals.

AH Plus endodontic sealer is considered the gold standard due to its excellent intratubular penetration properties, sealing capacity and non-interference of the endodontic sealer with the resin adhesive<sup>1</sup>. On the other hand, bioactive endodontic sealers have been developed to improve the quality of root canal obturation<sup>3</sup>, and bioceramic sealers such as Bio-C are becoming increasingly popular due to their ability to bond with the dentin surface and form hydroxyapatite<sup>4</sup>.

Fiberglass posts have similar physical properties to dentin, including modulus of elasticity, compressive strength, flexural strength and coefficient of thermal expansion, and provide advantages such as esthetics and biocompatibility<sup>5</sup>. Fiberglass posts can be cemented with conventional or self-adhesive resin sealers. The latter do not require pretreatment of the tooth surface, so the technique is simpler, thereby reducing sealing procedure time and risk of failure in the clinical steps<sup>6</sup>. The main cause of clinical failure of a fiberglass post is debonding at the dentin-sealer interface because of the degradation of the resin sealer-dentin bond over time<sup>7</sup>.

Various techniques have been reported in the literature to evaluate the bond strength of materials, including the conventional pull-out and push-out tests. Push-out tests provide better simulation of clinical conditions<sup>4</sup>, and enable evaluation of the effectiveness of adhesion in different regions of the canal, identifying precisely where the failures occur<sup>8</sup>. Given the limitations of the intraradicular adhesion procedure and the great challenge of achieving effective adhesion, it is important to evaluate the possible influence of different endodontic sealers on the bond strength of fiberglass posts to root dentin. Since there are few publications on the adhesion of bioceramic sealers, it was decided to compare them to AH Plus sealer, which is widely discussed in the literature due to its excellent properties.

The aim of this study was therefore to evaluate the bond strength between the fiberglass post cemented with self-adhesive resin sealer and the root dentin in teeth filled either with the bioceramic sealer Bio-C Sealer (Angelus, Londrina, Brazil) or with AH Plus (Dentsply Sirona, North Carolina, USA), using the push-out test. The null hypothesis tested was that the two sealers are similar both in terms of bond strength between the fiberglass post and the root dentin, and in terms of failure mode.

## MATERIALS AND METHOD

### Tooth selection and sample preparation

This study was approved by the Ethics Committee for Human Research of the São Leopoldo Mandic Research Institute (Approval number: 3.645.466). Forty-five single-rooted human teeth provided by the institutional Tooth Bank were divided into three groups. The sample size of 15 specimens per group, calculated by the Biostat 5.0 program, allowed a test power of 80% with a type I error probability of 0.05 and an effect size of 0.18.

The inclusion criteria were teeth with a single round canal of similar shape, size and diameter, with fully developed roots, with root curvatures of up to 15° according to Schneider<sup>9</sup> and an initial foramen diameter corresponding to a Kerr file #15. The round shape of the canals was verified radiographically, evaluating the symmetry between the vestibulolingual and mesiodistal distances. Exclusion criteria were teeth with calcifications, fissures, pathologic root resorption (internal, external or apical), root caries, previous endodontic treatment and root cracks visible under a surgical microscope with 10x magnification. The coronal section of each tooth was removed under water cooling with a double-sided diamond disk placed below the cemento-enamel junction. The root portion was standardized to an average length of 15 mm in the apical-cervical direction, measured with a conventional ruler and checked with a digital caliper. After preparation, the roots were immersed in distilled water and stored in an oven at 37 °C for rehydration until the tests were performed.

### Endodontic treatment and division into experimental groups

For root canal instrumentation, the working length was determined 1 mm short from the foramen, as a

with a #15 Kerr file tip was observed in the foramen under microscope at 12.5x magnification. All canals were prepared using a Logic 40./05 system (Bassi/Easy Equipamentos, Belo Horizonte, Brazil) driven by a Bassi iRoot Pro motor at 950 rpm and torque 4 N. After instrumentation, ultrasonically activated irrigation was performed to remove the smear layer, using an Irrisonic insert at 10% power and 30,000 Hz frequency, inserted 2 mm from the working length. Three 20-second cycles were performed with 2.5% NaOCl (5 mL), 17% EDTA (ethylenediaminetetraacetic acid) (5 mL), and 2.5% NaOCl (5 mL). The canals were irrigated with 5 mL of distilled water to remove the solutions. Finally, they were dried with an Ultradent suction cannula and capillary tip attached to a high-powered aspirator and 40-gage suction paper tips. A special random distribution program (<http://www.random.org>) was used to randomize the teeth into three groups (n=15) according to the endodontic sealer to be used. The protocol followed is shown in the flow chart in Fig. 1.

The groups were established as follows:

- Control group (GC): no endodontic sealer.
- Bio-C Sealer group (GBC): Bio-C Sealer, an endodontic sealer based on calcium silicate, which comes in a “ready-to-use” syringe.
- AH Plus group (GAH), AH Plus, an epoxy resin-based endodontic sealer in paste/paste presentation, which was mixed with a flexible spatula No. 24, so that the portions were the same for all samples.

The root canals were obturated with a single 40/05 guttaperchacone, 1 mm from the foramen. The sealers were placed in the canal with the gutta-percha cones and the thermoplastic tip and cold vertical compaction was performed. The teeth were then sealed with a temporary filling material (Coltosol, Coltene, Lezennes, France) and stored in an oven at 37 °C and 100% relative humidity for one week. The teeth were then sealed with a temporary filling material (Coltosol, Coltene, Lezennes, France) and stored in an oven at 37 °C and 100% relative humidity for one week.

### Preparation of the intra-radicular prosthetic space

Seven days after root canal obturation, the specimens were prepared for the insertion of fiberglass posts. The canals were partially exposed with Gates-

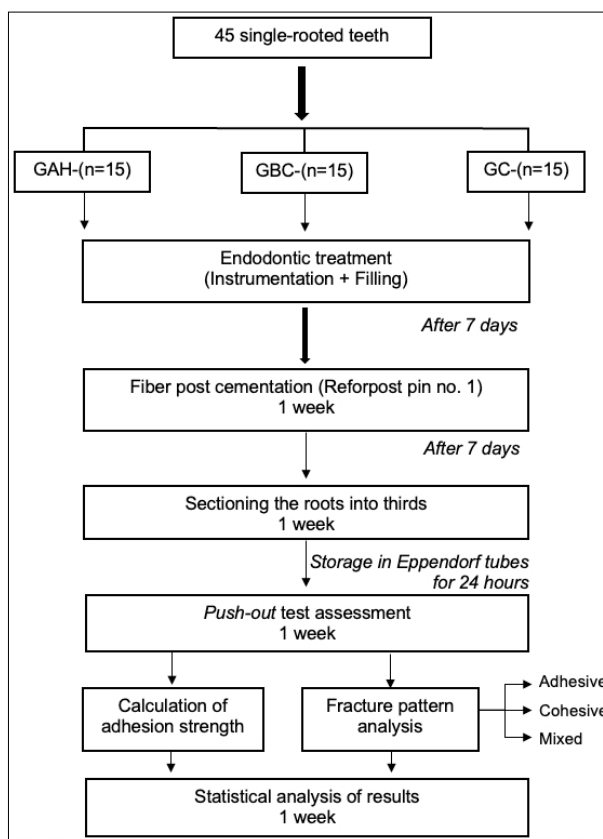


Fig. 1: Flow chart of the protocol followed in the experimental phase of this study.

Glidden #4 drills (Dentsply Sirona, Charlotte, USA) until an intraradicular length of 10 mm was reached. The preparation was then refined with Largo #3 burs (Dentsply Sirona) at low speed, following the fiber post manufacturer's instructions (Reforpost #1 post, Angelus, Londrina, Brazil) (Fig. 2).

After preparation for the post, the canals were irrigated with 1 mL of saline to remove debris and residual filling material, and then dried with absorbent paper points. The posts were immersed in 70% alcohol for one minute to remove oil and debris. The surfaces were treated with 37% phosphoric acid for 15 seconds, washed, and silanized for one minute, as recommended by the manufacturer, and then dried with a gentle stream of air.

The fiberglass posts were cemented with RelyX U200 self-adhesive resin sealer (3M do Brasil, Sumaré, Brazil), prepared according to the manufacturer's recommendations and inserted into the canal with a needle-shaped injection tip on a Maquira syringe. Immediately afterwards, the post was inserted into the root canal and the excess was removed with a microbrush. The cervical area of the roots was

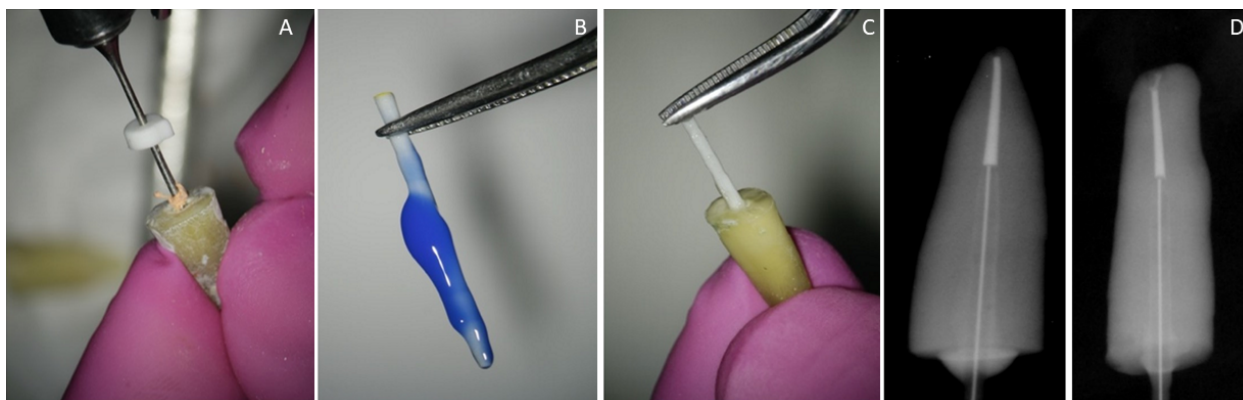


Fig. 2: Illustrative sequence of fiber post cementation and radiographic confirmation of adaptation

photoactivated for 60 seconds at a power of 1,200 milliwatts/cm<sup>2</sup> using a Radii-Cal photopolymerizer (Mosharraf & Zare<sup>10</sup>). After sealing the fiber posts, their fit was confirmed radiographically. The samples were stored in an environment at 100% humidity and temperature 37°C for seven days prior to testing.

The cemented root canal space was divided into cervical, middle and apical thirds. The roots were attached to a glass plate with sticky red wax. An IsoMet 1000 slicer (Buehler, Illinois, USA) with a 4"x12x1/2 EXTEC disk (Vilas-Boas et al.<sup>1</sup>) was used to cut 1.5 mm slices perpendicular to the long axis. The central slice of each third of the post space was selected and stored in an Eppendorf tube with distilled water for 24 hours.

### Mechanical extrusion shear test (push-out)

For the mechanical push-out test, each sample was positioned on a stainless-steel metal base on a universal testing machine (EMIC DL 2000, Instron Brasil Equipamentos Científicos Ltda, São José dos Pinhais, Brazil), equipped with a force of 2000 kgf. The disks were arranged so that the smaller base was facing upwards and the cervical side was in contact with the base. The force was applied from apical to coronal. A metal rod with a diameter of 1.0 mm and an active tip was attached to the clamp of the machine (Fig. 3) and positioned in the center of the fiber post.

The resistance test was performed at a rate of 1.0 mm/min until the pin and seal assembly was displaced or the specimen broke. The values were determined in Newtons, and the adhesive force of each disk was calculated by dividing the breaking force by the cross-sectional area in MPa. The adhesion area was calculated using the following formula:  $A = \pi(R +$

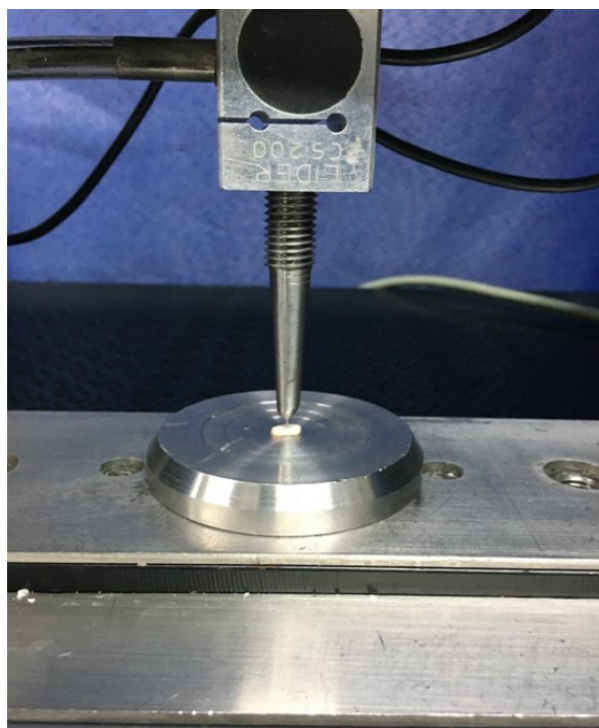


Fig. 3: Representative image of the slices adapted to the universal testing machine for the mechanical extrusion shear test (push-out).

$r) [h^2 + (R - r)^2]^{0.5}$ , where  $\pi$  is the fixed constant 3.14,  $R$  is the radius of the cemented post on the coronal side,  $r$  is the radius of the cemented post on the apical side and  $h$  is the thickness of the disk. The values obtained were tabulated and subjected to statistical analysis.

After push-out, the specimens were analyzed at 12.5x magnification in a DF Vasconcelos microscope to identify failure mode, which was classified as adhesive failure between dentin and sealer, cohesive failure in dentin, or mixed failure (Fig. 4).

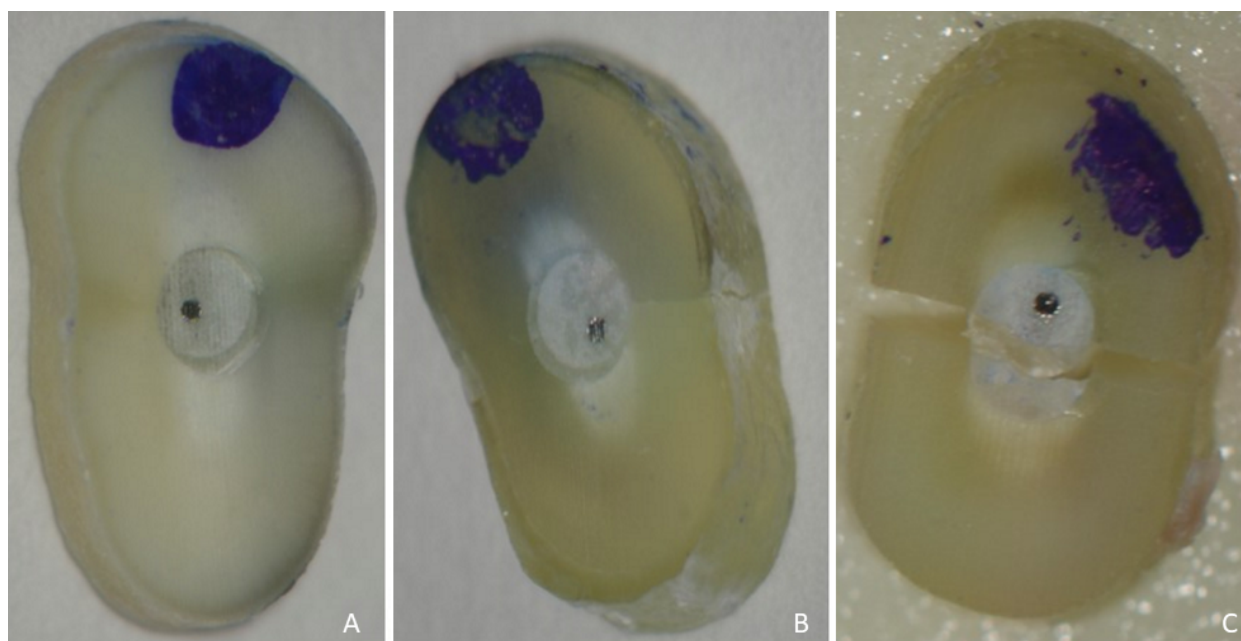


Fig. 4: Representative image of slices showing the types of gaps presented after the push-out test.

## RESULTS

The results were analyzed using the Biostat 5.0 program. The D'Agostino normality test showed non-normal behavior. The non-parametric Kruskal-Wallis (Student-Newman-Keuls) test was applied with a significance level of 1%.

Table 1 shows the medians and standard deviations of the push-out bond strength values according to endodontic sealer used. Fig. 5 shows the arithmetic means in a column diagram. In the cervical third, the highest bond strength occurred with the AH Plus sealer, with a significant difference only to Bioceramic sealer ( $p=0.0015$ ). In the middle third, there was no significant difference to any studied groups ( $p=0.1386$ ). In the apical third, the highest bond strength occurred with AH Plus sealer, with a significant difference to Bioceramic sealer ( $p=0.0014$ ) and control ( $p=0.0005$ ) (Table 1 and Fig. 5).

Comparing the same sealer in different thirds, there was significant difference only to Bioceramic sealer, which presented higher bond strength in the middle and apical thirds, with a significant difference in relation to the cervical third ( $p<0.05$ ). The other comparisons did not present statistically significant differences ( $p>0.05$ ) (Table 1 and Fig. 5).

Regarding failure types, no differences were observed among the endodontic sealers BC (Bioceramic Sealer), AH (AH Plus Sealer), and C

**Table 1. Medians, interquartile deviations and statistical analysis of the bond strength (MPa) of the endodontic sealers BC (Bioceramic Sealer), AH (AH Plus Sealer) and C (control group) in the Cervical (C), Middle (M) and Apical (A) thirds.**

|            | BC                        | AH                        | C                           | (p)             |
|------------|---------------------------|---------------------------|-----------------------------|-----------------|
| <b>C</b>   | 1.70 (1.90) <sup>a1</sup> | 3.95 (3.04) <sup>a2</sup> | 2.30 (2.62) <sup>a1,2</sup> | <b>&lt;0.05</b> |
| <b>M</b>   | 3.34 (2.38) <sup>b1</sup> | 4.29 (3.63) <sup>a1</sup> | 4.83 (3.54) <sup>a1</sup>   | <b>&gt;0.05</b> |
| <b>A</b>   | 4.11 (1.97) <sup>b1</sup> | 5.90 (3.12) <sup>a2</sup> | 2.93 (2.46) <sup>a1</sup>   | <b>&lt;0.01</b> |
| <b>(p)</b> | <b>&lt;0.05</b>           | <b>&gt;0.05</b>           | <b>&gt;0.05</b>             |                 |

Different letters in the vertical direction and different numbers in the horizontal direction denote statistically significant differences.

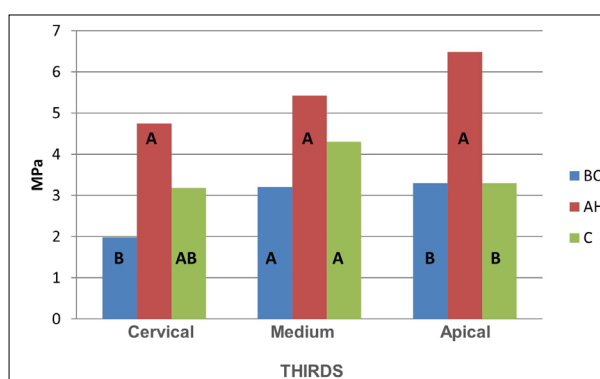


Fig. 5: Arithmetic means of the bond strength (MPa) of the endodontic BC (Bioceramic), AH (AH Plus Sealer) and C (control group) in the Cervical (C), Middle (M) and Apical (A) thirds. Legend: BC: Bioceramic Sealer; AH: AH Plus Sealer and C: control group.

Different letters denote statistically significant differences.

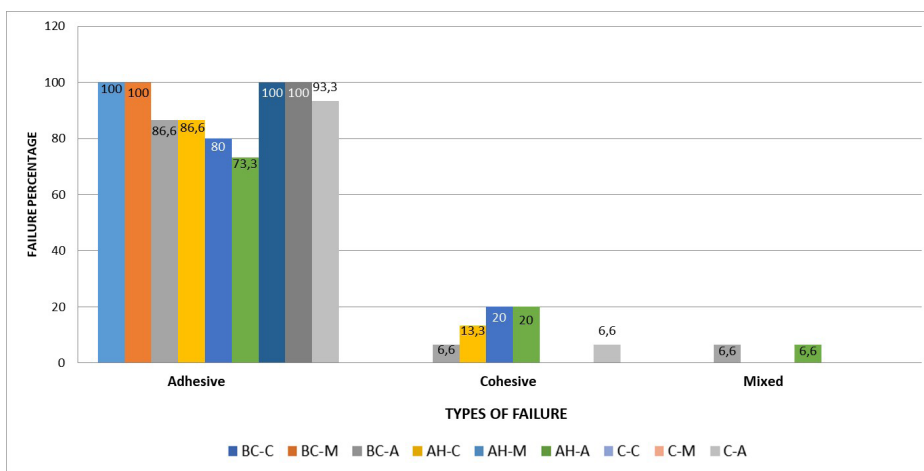


Fig. 6: Percentage of fracture types presented by endodontic BC (Bioceramic), AH (AH Plus) and C (control group) in the Cervical (C), Middle (M) and Apical (A) thirds.

Legend: BC-C: bioceramic cervical third; BC-M: bioceramic middle third; BC-A: bioceramic apical third; AH-C: AH Plus cervical third; AH-M: AH Plus middle third; AH-A: AH Plus apical third; C-C: control cervical third; C-M: control middle third; C-A: control apical third.

**Table 2. Medians, interquartile deviations and statistical analysis of the types of failure presented by the endodontic sealers BC (Bioceramic Sealer), AH (AH Plus Sealer) and C (control group) in the Cervical (C), Middle (M) and Apical (A) thirds.**

|            | BC              | AH              | C               | (p)             |
|------------|-----------------|-----------------|-----------------|-----------------|
| <b>C</b>   | 1.00 (0.00)A    | 1.00 (0.00)A    | 1.00 (0.00)A    | <b>&gt;0.05</b> |
| <b>M</b>   | 1.00 (0.00)A    | 1.00 (0.00)A    | 1.00 (0.00)A    | <b>&gt;0.05</b> |
| <b>A</b>   | 1.00 (0.00)A    | 1.00 (0.50)A    | 1.00 (0.00)A    | <b>&gt;0.05</b> |
| <b>(p)</b> | <b>&gt;0.05</b> | <b>&gt;0.05</b> | <b>&gt;0.05</b> |                 |

Failure type scores: Adhesive 1; Cohesive: 2 and Mixed: 3. Equal letters and numbers in the vertical and horizontal directions denote absence of statistically significant differences.

(control group) in the cervical, middle, and apical thirds (Fig. 6 and Table 2).

## DISCUSSION

The null hypothesis regarding bond strength was rejected because the different endodontic sealers influenced the bond strength of the fiberglass post to the root dentin in the cervical and apical thirds of the post. The null hypothesis regarding failure type was accepted because there was no difference between the tested groups. In the present study, the control group was treated only with gutta-percha, without endodontic sealer, as frequently reported in the literature<sup>1,4,8,11,12</sup>, so that the endodontic sealer was the only variable between the control group and the experimental groups.

Fiberglass posts are often used in the rehabilitation

of endodontically treated teeth. The main factors that affect the retention of posts are dimensions (length, diameter), shape (conical, cylindrical), type of surface (fluted, threaded or smooth), intracanal space preparation, type of sealer, and the skill of the practitioner<sup>11</sup>. The procedures in this study were performed by a single experienced operator. Reforpost fiber post No. 1 (Angelus) is made of fiberglass, pigmented epoxy resin and a stainless steel filament, has a parallel shape with apical taper and serrated surface. It was selected because it contains a steel filament which enables radiographic visualization to verify post positioning in the intraradicular space.

The intraradicular length used was 10 mm, respecting the 5 mm wide space of the canal obturation, as reported by Baena et al.<sup>13</sup>, Vilas-Boas et al.<sup>1</sup> and Chen et al.<sup>14</sup>.

The push-out (or extrusion shear test) was used in this study because it provides great precision in the measurement of sealer resistance, evenly distributed stress, and low variability in the mechanical test, thereby enabling evaluation of the differences in bond strength between the root thirds. It is therefore recommended for determining the bond strength of fiberglass posts to intraradicular dentin<sup>10,11,15</sup>.

The irrigation solution used is one of the factors that may affect the adhesion of the fiberglass post to the root dentin. Chemical adjuvants in the irrigating solution, such as NaOCl and EDTA in different concentrations, are commonly used and

widely accepted for removing organic and inorganic residues from the root canals. While the halogenated solution promotes the dissolution of organic tissue, the chelating agent acts on the inorganic portion of the smear layer<sup>10,15,16</sup>. Thus, to simulate the clinical mode, we used 2.5% NaOCl and 17% EDTA as rinsing solutions.

The relationship between the adhesion of fiberglass posts and the type of endodontic sealer used in root canal filling has been extensively discussed in the literature because some endodontic sealers can have negative effects on resin sealer polymerization, thereby compromising the longevity of restorations held in place by intraradicular posts<sup>1,2,4,8,10,11,17</sup>.

Vilas-Boas et al. found no difference in the bond strength of the fiber post when different sealers were tested<sup>1</sup>. However, the present study found in the apical third of the post space, bond strength was lower in the control group than in the AH Plus group, suggesting that the AH Plus sealer does not impair post adhesion, and may promote better retention. This may be due to the presence of residual resin from the AH Plus endodontic sealer on the canal walls, which could foster interaction with the resin sealer, as shown by Mosharraf & Zare<sup>10</sup>.

Bioceramic endodontic sealers based on calcium silicate are recognized for their biocompatibility and ease of use due to their premixed form<sup>14</sup>. In this study, sealers were used with a gutta-percha cone to standardize methodology. There are few studies on the adhesion of fiber posts with bioceramic sealers, which have only been introduced recently. There is no consensus on the timing for post space preparation after obturation. Rosa et al.<sup>17</sup> found no effect of timing on post adhesion, in agreement with Menezes et al.<sup>18</sup> and Vilas-Boas et al.<sup>1</sup>. However, other studies, such as Yuanli et al.<sup>2</sup>, noted lower adhesion with immediate cementation. The current study prepared the posts after seven days, following Ozcan et al.<sup>8</sup> and Bengoa et al.<sup>12</sup>, to ensure complete curing.

Resin cements are used for cementing fiberglass posts, and the literature contains numerous studies evaluating their effects on the bond strength of fiberglass posts. They can be divided into conventional cements (which were preceded by adhesive systems) and self-adhesive cements. Bengoa et al.<sup>12</sup>, Yuanli et al.<sup>2</sup>, and the current study used the self-adhesive sealer U200 to install fiberglass posts, but other studies such as Dibaji et

al.<sup>4</sup> and Vilas-Boas et al.<sup>1</sup> used conventional sealers. In contrast to the adhesion mechanism of conventional resin sealers, self-adhesive ones do not form a pronounced hybrid layer or a resin tag. There is surface interaction with the dental substrate due to the chemical interaction with the calcium of the hydroxyapatite<sup>7,13</sup>. Although self-adhesive sealers have been reported to have lower adhesion values than conventional ones by Chávez-Lozada, Urquía-Morales<sup>6</sup>, they were proposed by Sarkis-Onofre et al.<sup>19</sup> in a systematic review for better retention of fiberglass posts in root canals. They are easy to handle, have dimensionally stable mechanical retention properties, have better physical properties than conventional ones, are moisture tolerant, have good compressive strength and microhardness, and are less sensitive to technique due to their one-step nature, as they do not require pretreatment with bonding agents on the tooth surface, which makes their clinical application attractive<sup>7</sup>. The self-adhesive sealer RelyX U200 was chosen for fiber post adhesion in this study.

Although there is no consensus on the best method for removing debris from the root canal post space, treating dentin with NaOCl and EDTA may enhance bond strength between resin sealer and dentin. Baena et al.<sup>13</sup> found that bond strength increased when dentin was treated with phosphoric or polyacrylic acid before sealing. Phosphoric acid removes the smear layer and demineralizes the dentin, enhancing resin infiltration and micromechanical interlocking. Polyacrylic acid partially removes the smear layer and creates a surface that enhances chemical interaction with resin sealers. However, conditioning with 17% EDTA showed no adhesion benefit, in agreement with Moura et al.<sup>16</sup>, who found no influence of EDTA on RelyX U200 adhesion. EDTA, a chelating agent, lowers calcium levels and does not enhance micromechanical bonding. Studies by Martinho et al.<sup>20</sup> and Chen et al.<sup>14</sup> found ultrasound activation ineffective in improving smear layer removal and bond strength. Ferreira et al.<sup>15</sup> highlighted the importance of cleaning dentin walls under microscopy for better adhesion. Given the controversy, the current study chose not to pretreat the dentin, using only a saline solution rinse, as the self-adhesive sealer does not require pretreatment. Some studies compare fiber post adhesion by root third regions<sup>2,4,8,12,21</sup>, as was done in the current study. We observed greatest adhesion in the apical

third, followed by the middle and cervical thirds for both sealers evaluated, although the difference was significant only in the Bioceramic group. This agrees with Jha & Jha<sup>21</sup>, who attributed the difference to stronger reamer wear in the apical thirds than in the cervical thirds during canal preparation for the post. This may eliminate sealer residues in the dentin and facilitate interaction with the sealing agent.

Boing et al.<sup>22</sup> linked the effectiveness of RelyX U200 sealer to its reaction with hydroxyapatite, noting that adhesion was better in the apical third due to more available calcium and increased contact with canal walls supporting both chemical and mechanical retention. In the current study, the lower adhesion in the cervical third with bioceramic sealer (GBC) compared to AH Plus (GAH) may result from the manufacturer's recommended insertion method, as gutta-percha cones may reduce sealer penetration into tubules and thus, also adhesion.

In contrast, studies such as Menezes et al.<sup>18</sup> and Soares et al.<sup>23</sup> detected greatest adhesion in the cervical third, followed by the middle and apical thirds. This was explained by the fact that there are more dentinal tubules and larger diameters in the cervical third than in the others. In addition, light attenuation by the fiber post decreases with increasing root depth.

In the literature, the lowest values are reported in the apical region, which is associated with the difficulty of the adhesion protocol (several steps for dentin hybridization and the type of resin sealer used), the presence of gutta-percha and sealer residues, and the lack of light that fully reaches the interior of the root canals, which affects the adequate resin sealer curing<sup>20</sup>.

Sometimes there is no significant difference between the thirds evaluated, as reported by Ozcan et al.<sup>8</sup> and Baena et al.<sup>13</sup>. These results are confirmed by Pereira et al.<sup>24</sup>, who found that the region of the post space did not influence the dual resin sealer conversion.

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In other words, this homogeneous behavior is explained by the dual curing of the sealer, which enables polymerization where the light does not reach.

This study found that endodontic sealer type influences post adhesion in the cervical and apical thirds, with AH Plus showing stronger adhesion than bioceramic sealer. These findings agree with Vilas-Boas et al.<sup>1</sup>, Dibaji et al.<sup>4</sup>, and Bengoa et al.<sup>12</sup>. However, Ozcan et al.<sup>8</sup>, Rosa et al.<sup>17</sup>, and Reyhani et al.<sup>11</sup> found no adhesion differences with calcium silicate or MTA-based sealers, possibly due to the resin component in MTA Fillapex. Yuanli et al.<sup>2</sup> also found no difference between AH Plus and bioceramic sealers with immediate post cementation. No significant differences were found in middle third adhesion, in agreement with Bengoa et al.<sup>12</sup>, though this study attributes it to the non-conical Reforpost design rather than to canal morphology.

Failure mode analysis showed no differences between sealers, with prevalent adhesive failures between dentin and sealer, explained by the high C-factor and reduced light intensity in root dentin<sup>17</sup>. The study found differences in bond strength between root dentin and fiberglass posts when self-adhesive resin sealer was used. Bioceramic sealer may compromise post adhesion. Most research, including this study, involves in vitro methods, presenting limitations. Further research is recommended due to the clinical relevance of the issue and potential for treatment failure.

#### CONCLUSION

The results suggest that the bond strength of fiberglass posts cemented with U200 self-adhesive resin sealer is affected less by AH Plus than by the bioceramic sealer. The most common type of failure was adhesive type at the interface between the sealer and the root dentin, regardless of the endodontic sealer used.

#### CONFLICT OF INTERESTS

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

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# Smile-related oral characteristics in vietnamese students

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## ABSTRACT

**Aim:** We evaluated various oral features related to the smile among Vietnamese students. Additionally, we identified gender differences in the oral features and the prevalence of attractive smiles. **Materials and Method:** We recruited 490 Vietnamese students with 18-29 years old and recorded their dynamic smiles. We determined the prevalence of the qualitative smile morphological features and the mean and standard deviation of the quantitative features, namely the buccal corridor space, the smile index, and dynamic smile symmetry. **Results:** There were diverse smile-related oral characteristics among the participants. The most common anterior smile line was “average” (42.6%), and “parallel” was the most prevalent smile arc (43.1%). We observed a “downward” upper lip curvature in 55.9% of the participants, and the “second premolar” was the most frequently displayed posterior tooth (58.6%). Most of the participants (79.2%) showed no contact between the upper teeth and the lower lip while smiling. The mean buccal corridor space was  $0.59 \pm 0.47$  cm, the smile index was  $5.85 \pm 1.48$ , and dynamic smile symmetry was  $1.01 \pm 0.06$ . There were significant gender differences for several smile-related characteristics. **Conclusions:** Women more often display a favorable smile arc and more teeth when smiling, while men are more likely to have an attractive anterior smile line and upper lip curvature. These insights are vital to tailor effective aesthetic treatments.

**Keywords:** smiling - dental care - surgical procedures - students

## Características orales relacionadas con la sonrisa en estudiantes vietnamitas

## RESUMEN

**Objetivo:** Evaluamos diversas características bucales relacionadas con la sonrisa entre estudiantes vietnamitas. Además, identificamos las diferencias de género en las características orales y en la prevalencia de sonrisas atractivas.

**Materiales y Método:** Se reclutaron 490 estudiantes vietnamitas de entre 18 y 29 años y se registraron sus sonrisas dinámicas. Se determinó la prevalencia de las características morfológicas cualitativas de la sonrisa, así como la media y la desviación estándar de las características cuantitativas, a saber: el espacio del corredor bucal, el índice de sonrisa y la simetría de la sonrisa dinámica.

**Resultados:** Se observaron diversas características orales relacionadas con la sonrisa entre los participantes. La línea de sonrisa anterior más común fue la “media” (42,6%), y el arco de sonrisa más prevalente fue el “paralelo” (43,1%). Se observó una curvatura del labio superior “hacia abajo” en el 55,9% de los participantes, y el “segundo premolar” fue el diente posterior mostrado con mayor frecuencia (58,6%). La mayoría de los participantes (79,2%) no presentaron contacto entre los dientes superiores y el labio inferior al sonreír. El espacio medio del corredor bucal fue de  $0,59 \pm 0,47$  cm, el índice de sonrisa fue de  $5,85 \pm 1,48$  y la simetría de la sonrisa dinámica fue de  $1,01 \pm 0,06$ . Se encontraron diferencias significativas entre hombres y mujeres en varias características relacionadas con la sonrisa.

**Conclusiones:** Las mujeres muestran con mayor frecuencia un arco de sonrisa favorable y exhiben más dientes al sonreír; mientras que los hombres tienden a presentar una línea de sonrisa anterior y una curvatura del labio superior más atractivas. Estos hallazgos son fundamentales para personalizar tratamientos estéticos efectivos.

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## INTRODUCTION

Smiles are universally recognized as a key expression of happiness, but their morphological features can differ significantly across various ethnic groups. Much of the existing research on smile dynamics and aesthetics has been concentrated on Caucasian populations, resulting in a restricted understanding of how ethnic differences shape the expression of smiles and the perceptions of beauty. This narrow focus complicates efforts to generalize findings from European-American samples to Asian populations, such as Vietnamese individuals, due to distinct morphological and cultural factors. Consequently, there is a crucial need for research that explores smile characteristics within diverse ethnic groups to achieve more accurate and culturally pertinent insights<sup>1,2</sup>.

In recent years, researchers have begun to address the ethnic gap by examining the smile characteristics of Chinese, Japanese and Indian populations<sup>3-8</sup>. These studies have yielded significant findings, such as variations in the smile curve, the upper lip arch, and teeth exposure. For example, research on Chinese individuals has highlighted distinctions in the types of smiles, with a predominance of commissure and cuspid smiles<sup>7</sup>. Similar variations in smile line and smile arc in a smile have been highlighted by studies on Indian smiles<sup>5,6</sup>. In Vietnam, researchers have begun to identify certain characteristics of smile aesthetics. For example, Pham<sup>9</sup> observed common traits of smile lines, a rising curvature of upper lip, and a parallel smile arc. Moreover, smiles often expose teeth up to the second premolar. Nevertheless, the current research was constrained by a small size of samples and did not offer a thorough overview of the morphological smile features, particularly regarding the interplay between the mouth and face. Understanding the complex nature of smiles requires an assessment of various features. Together, these components add to a smile's distinct beauty and expressiveness. There is an urgent need for region-specific analyses that consider the cultural and morphological features of smiles, given the significance of ethnic differences in aesthetic perceptions. There is a lack of data about smiles and their related oral features in Vietnam. To the best of our knowledge, there have been no comprehensive assessments on the smile-related oral characteristics on Vietnamese subjects of a wide range of ages. This endeavor is necessary to generate a comprehensive

database regarding the smile morphological features and the related oral factors of Vietnamese people. This information would support the establishment of treatment plans to improve the smile design and to perform future research on the beauty of Vietnamese smiles.

Given the aforementioned gaps in the literature, we conducted a comprehensive investigation into the morphology of smiles in Vietnamese participants of different ages. We evaluated various oral aspects of smiles among students at Vietnam National University, Ho Chi Minh City (VNU-HCM). Then, we determined gender differences in these smile features and the prevalence of attractive smiles. Our findings should help improve personalized treatment planning and benefit the larger field of facial aesthetics.

## MATERIALS AND METHOD

### Sampling and selection criteria

The sampling and smile recording processes are shown on Figure 1. A comprehensive dental check-up process was conducted to ensure the selection of a suitable study sample. Initially, the participants underwent a detailed oral examination conducted by dentists to assess their eligibility. This included a detailed oral examination and assessment of dental history, ensuring the presence of the necessary teeth and proper occlusion. Four hundred and ninety subjects (323 men and 167 women) Vietnamese students with the mean age of  $21.04 \pm 1.79$  (18-29 years old) studying at VNU-HCM participated in this study. Criteria for choosing participants were as follows: (1) none of the eight anterior maxillary teeth were missing, there was mild or no crowding and no other malposition conditions; (2) missing no more than four maxillary and mandibular teeth (excluding the wisdom teeth); (3) Angle class I molar occlusion; (4) no record of orthodontic surgery or cosmetic surgery; and (5) no maxillofacial trauma, deformities, or lip abnormalities. The study process is illustrated in (Fig. 1). This research was approved by the Human Subjects Ethics Board of the University (number 256/DHYD-HDDD).

The students who met the sampling criteria are required to take part in a demographic survey to record their names, ages, genders, education, and jobs. The participants had their teeth scaled 1 week before their smiles were recorded. The participants

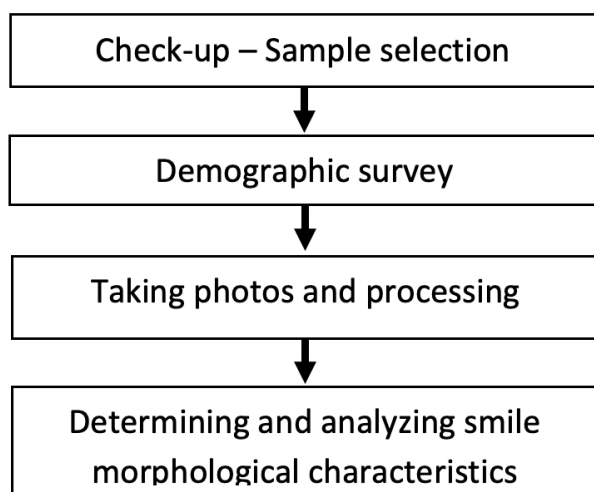


Fig. 1: The procedure used for dynamic smile recording

were instructed to sit up straight with their arms hanging naturally and their eyes looking straight at the camera. The head was adjusted so that the Frankfort plane was parallel to the floor throughout the imaging procedure. A ruler was placed with the smallest division of 1 mm close to the right side of the participant's face so that the distance from the lens to the ruler and to the central incisor was equal.

The entire imaging process was standardized and the same for all imaging sessions. A Canon EOS 6D digital camera and accompanying flash system was placed on a camera tripod 150 cm from the participant's upper lip to the closest point of the camera. The background was plain white. The center of the camera lens was placed above the occlusal plane, and the shooting direction was perpendicular to the sagittal plane. Two images were recorded for each patient: an image of the mouth when smiling naturally, and an image of the mouth at rest. The camera was connected directly to the computer to control image quality. A satisfactory image was high resolution, was not blurred, and had sufficient brightness and realistic colors. All photos were taken by a single trained photographer.

The captured images were saved to a computer as JPEG files and edited by Adobe Photoshop CS6 software. They were adjusted to remove the effect of facial features and skin colors on aesthetic evaluation. Finally, these images were standardized in a size of 5 × 3 inches, black and white color, and 70 dpi, and saved as JPEGs. These images were copied into PowerPoint to perform the evaluation. All images were evaluated by the same previously

trained dentist to assess smile characteristics according to the following criteria.

1. There were 4 types of anterior smile line based on the visible teeth and gingiva<sup>3</sup>. The first type is very high, which indicates > 2-mm marginal gingiva visible. The second type is high which indicates 0-2-mm marginal gingiva while the third type is average which indicates only the gingiva embrasure. The last type was low, which was impossible to see the gingiva.
2. The smile curve<sup>3</sup> is determined based the correlation between the line drawn along the incisal edges of the maxillary central incisors and the superior border of the lower lip. There are 3 types, including a parallel smile arc, a straight smile arc, and a reverse smile arc.
3. The upper lip curvature<sup>3</sup> describes the morphology of the inferior border of the upper lip based on the relationship between the corner of the mouth and the center of the inferior border of the upper lip. There are 3 types: a straight lip curvature, a downward lip curvature, and an upward lip curvature.
4. The most posterior tooth displayed when smiling<sup>3</sup> is divided as up to the canine, up to the first premolar, up to the second premolar, up to the first molar, or up to the second molar.
5. The relationship between the upper teeth and the lower lip<sup>1</sup> has three categories: not touch, touching, and slightly covering
6. For the buccal corridor space<sup>10</sup>, the black area that forms at the corners of the mouth when smiling horizontally is measured.
7. The smile index<sup>3</sup> is the ratio of the horizontal distance and the vertical distance. It is determined based on four reference points in (Fig. 2) as  $[(1 - 2) / (3 - 4)]$ .
8. The dynamic smile symmetry<sup>3</sup> refers to the uniformity of movement of the bilateral outer commissure in the horizontal and vertical direction. It is determined based on the four reference points in (Fig. 2) as  $[(1 - 3) + (1 - 4)] / [(2 - 3) + (2 - 4)]$ .

### Data analysis

The software to be used in the current study was SPSS Statistics version 23.0 for Windows. The prevalence of the qualitative smile morphological features and the mean ± standard deviation (SD) of



Fig. 2: The reference points used to calculate the smile index and dynamic smile symmetry

the quantitative features was determined. Gender differences were determined with the chi-squared test (for the qualitative features) and the t-test (for the quantitative features).

### Research ethics

The current study follows the regulations of biomedical research ethics. The students (and their families) were informed about their oral health status. All of the information collected is confidential and for research purposes only.

## RESULTS

### Smile-related oral characteristics

Table 1 shows the qualitative smile-related oral characteristics. For the anterior smile line, the majority of individuals fell into the average category (42.6%), followed by the low (32.7%), high (13.3%), and very high (11.4%) categories. Regarding the smile arc pattern, parallel (43.1%) was most prevalent, followed by straight (26.1%) and reverse (30.8%). The majority of participants had a downward curvature (55.9%), while 33.9% displayed a straight upper lip, and 10.2% had an upward curvature. For most posterior tooth displayed when smiling, the second premolar was the most frequent (58.6%), followed by the first molar (20.2%) and the first premolar (20.0%). The second molar and canine were rarely the most posterior tooth visible, each accounting for less than 1% of the total sample. Finally, when evaluating the relationship between the upper and lower lip, the majority of the participants (79.2%) exhibited no contact between their upper and lower lip. A smaller proportion demonstrated touching (17.1%), while a few participants had slightly covering (3.7%).

**Table 1: The qualitative smile-related oral characteristics**

| Feature                                                |                   | N   | %    |
|--------------------------------------------------------|-------------------|-----|------|
| Anterior smile line                                    | Low               | 160 | 32.7 |
|                                                        | Average           | 209 | 42.6 |
|                                                        | High              | 65  | 13.3 |
|                                                        | Very high         | 56  | 11.4 |
| Smile arc                                              | Reverse           | 151 | 30.8 |
|                                                        | Parallel          | 211 | 43.1 |
|                                                        | Straight          | 128 | 26.1 |
| Upper lip curvature                                    | Upward            | 50  | 10.2 |
|                                                        | Downward          | 274 | 55.9 |
|                                                        | Straight          | 166 | 33.9 |
| Most posterior tooth displayed                         | First premolar    | 98  | 20   |
|                                                        | Second premolar   | 287 | 58.6 |
|                                                        | First molar       | 99  | 20.2 |
|                                                        | Second molar      | 2   | 0.4  |
|                                                        | Canine            | 4   | 0.8  |
| Relationship between the upper teeth and the lower lip | Touching          | 84  | 17.1 |
|                                                        | Not touching      | 388 | 79.2 |
|                                                        | Slightly covering | 18  | 3.7  |

Abbreviation: N = Sample size

**Table 2: The quantitative smile-related characteristics**

| Measure                | Minimum | Maximum | Mean | Standard deviation |
|------------------------|---------|---------|------|--------------------|
| Buccal corridor space  | 0       | 2       | 0.59 | 0.47               |
| Smile index            | 2.50    | 13.40   | 5.85 | 1.48               |
| Dynamic smile symmetry | 0.85    | 1.21    | 1.01 | 0.06               |

Table 2 presents the data for the quantitative smile-related characteristics. The mean  $\pm$  SD was  $0.59 \pm 0.47$  for the buccal corridor,  $5.85 \pm 1.48$  for the smile index, and  $1.01 \pm 0.06$  for dynamic smile symmetry. Figures 3- present representative examples of smile patterns analyzed in this study. Consistent with the criteria proposed by Pham<sup>9</sup>, (Fig. 3) illustrates a prototypical smile that encompasses all four key characteristics contributing to smile attractiveness. (Figs. 4,5) demonstrate specific aesthetic features considered attractive that appears more often in male and female smiles, respectively, as identified through the current analysis.

#### Gender differences for smile-related characteristics

Table 3 showed that the results were significant ( $p < 0.05$ ) gender differences and dynamic smile symmetry.

#### DISCUSSION

We have provided a comprehensive analysis of smile-related oral characteristics among Vietnamese students, revealing significant variations in the anterior smile line, the smile arc, the upper lip curvature, and other key features. The most prevalent anterior smile line was average (42.6%), while the



Fig. 3: A smile exhibiting all four features associated with aesthetic attractiveness, including an average or high anterior smile line, a parallel smile arc, an upward curvature of the upper lip, and visibility of the second premolar as the most posterior tooth displayed



Fig. 4: A smile of a female participant demonstrating two of the four established features of smile attractiveness, namely a parallel smile arc and visibility of the second premolar as the most posterior tooth displayed



Fig. 5: Smile of a male participant exhibiting two of the four recognized features of smile attractiveness, specifically an average or high anterior smile line and an upward curvature of the upper lip.

parallel smile arc was the most common (43.1%). Notably, the majority of the participants exhibited a downward upper lip curvature (55.9%), and the second premolar was the most frequently displayed posterior tooth (58.6%).

The distribution of smile characteristics greatly contributed to the typical aesthetic preferences and anatomical features of the Vietnamese population. The predominance of the average anterior smile line and the parallel smile arc are similar to research on the Germany population<sup>1</sup>. According to a previous study of Machado<sup>4</sup>, people with a parallel smile arc tend to appear younger, happier, and more beautiful than those with a nonparallel smile arc. This type of smile arc is also known as the “consonant” smile arc, and according to Krishnan et al<sup>6</sup>, orthodontists should not intervene with this type of smile; rather, they should use the right bracket positioning to create it. The downward upper lip curvature, seen in over half of our participants, contrasts with the more common upward curvature observed in Western populations in Germany<sup>1</sup> and Spain<sup>2</sup>. This difference

**Table 3: Gender differences in the smile-related oral characteristics**

| Feature                                                |                   | Gender      |              | Statistical analysis |
|--------------------------------------------------------|-------------------|-------------|--------------|----------------------|
|                                                        |                   | Male N (%)  | Female N (%) | P*                   |
| Anterior smile line                                    | Low               | 125 (38.7)  | 35 (21)      | <0.001               |
|                                                        | Average           | 151 (46.7)  | 58 (34.7)    |                      |
|                                                        | High              | 31 (9.6)    | 34 (20.4)    |                      |
|                                                        | Very high         | 16 (5)      | 40 (24)      |                      |
| Smile arc                                              | Reverse           | 100 (31)    | 51 (30.5)    | <0.001               |
|                                                        | Parallel          | 115 (35.6)  | 96 (57.5)    |                      |
|                                                        | Straight          | 108 (33.4)  | 20 (12)      |                      |
| Upper lip curvature                                    | Downward          | 194 (60.1)  | 80 (47.9)    | 0.014                |
|                                                        | Straight          | 95 (29.4)   | 71 (42.5)    |                      |
|                                                        | Upward            | 34 (10.5)   | 16 (9.6)     |                      |
|                                                        | Canine            | 4 (1.2)     | 0 (0)        |                      |
| Most posterior tooth displayed when smiling            | First premolar    | 64 (19.8)   | 34 (20.4)    | 0.031                |
|                                                        | Second premolar   | 177 (54.8)  | 110 (56.9)   |                      |
|                                                        | First molar       | 76 (23.5)   | 23 (13.8)    |                      |
|                                                        | Second molar      | 2 (0.6)     | 0 (0)        |                      |
| Relationship between the upper teeth and the lower lip | Touch             | 48 (14.9)   | 36 (21.6)    | 0.063                |
|                                                        | Not touch         | 260 (80.5)  | 128 (76.6)   |                      |
|                                                        | Slightly covering | 15 (4.6)    | 3 (1.8)      |                      |
| Total                                                  |                   | 323 (100)   | 167 (100)    |                      |
| Measure                                                |                   | Mean ± SD   | Mean ± SD    | P**                  |
| Buccal corridor space                                  |                   | 0.6 ± 0.54  | 0.57 ± 0.29  | 0.454                |
| Smile index                                            |                   | 5.81 ± 1.57 | 5.93 ± 1.28  | 0.409                |
| Dynamic smile symmetry                                 |                   | 1.01 ± 0.07 | 0.99 ± 0.05  | 0.001                |

Abbreviations: N = sample size; SD = standard deviation  
 \* Pearson  $\chi^2$  test  
 \*\* t-test.

emphasizes the importance of considering ethnic-specific characteristics in dental aesthetics, as applying a Western-centric standard to Vietnamese patients may not achieve optimal results.

In the Vietnamese context, Pham<sup>9</sup> outlined some characteristics of smile aesthetics, including a high or average smile line, an upward upper lip curvature, and a parallel smile arc. Compared with this previous study, our study's strength lies in its comprehensive

evaluation of smile characteristics across a broader sample size (490 participants compared with 200) and its focus on additional features. Unlike that previous study, which primarily addressed perceptual differences, our study provides a more detailed analysis, revealing that a significant portion of Vietnamese individuals have a downward upper lip curvature and display up to the second premolar when smiling. Because the upper lip curvature cannot be changed, it is more difficult for patients with a downward lip curvature to achieve their ideal smile. Thus, these findings suggest that ethnic-specific characteristics are crucial for understanding and improving smile aesthetics in the Vietnamese population.

Our findings align with previous research conducted on different populations, emphasizing the importance of ethnic-specific studies in understanding smile aesthetics. For example, Wang<sup>3</sup> and Liang<sup>7</sup> investigated Chinese smiles and highlighted variations in the smile arc and upper lip curvature, findings that are in line with our observations regarding the parallel smile arc. However, we found that a downward upper lip curvature was the most popular in our population, which is upward in the study of Wang<sup>3</sup> and straight in the one of Liang<sup>7</sup>. Hence, ethnic and cultural factors influence smile characteristics.

Regarding the smile line, in Germany, researchers found that the majority of patients (52%) had an average smile line, followed by 38% with a high smile line and 10% with a low smile line<sup>1</sup>. This suggests a predominant tendency towards average smile lines within the German population. In Spain, there was an even stronger prevalence of average smile lines, with 84.3% of patients exhibiting this characteristic, while only 8.6% had a low smile line and 7.1% had a high smile line, indicating a clear preference or genetic inclination toward an average smile line in the Spanish population<sup>2</sup>. In contrast, Pham<sup>9</sup> showed a different distribution, with nearly half of the patients (49.5%) having a high and very high smile line, 33% with an average smile line and 17.5% with a low smile line. The higher dominance of a high smile line suggests distinct aesthetic or anatomical differences prevalent in the Vietnamese population. Our findings are different: 42.6% of patients presented an average smile line, 32.7% a low smile line, and 24.7% a high smile line. Compared with the previous Vietnamese study, we

found a noticeable increase in an average smile line and a reduction in a high smile line. This feature is very important in treatment of patients to avoid excessive gingival display that can affect the overall smile aesthetics.

Considering the most posterior tooth displayed when smiling, our findings align with previous studies, reflecting similar trends and distributions of dental display, which was mostly up to the second premolar. However, we revealed a more diverse range of the most posterior tooth displayed when smiling, including up to the second molar and even the canine<sup>1, 2, 9, 11</sup>. Moreover, most of the participants had upper teeth not touching their lower lip. These findings were similar to that reported in previous studies: This dentolabial relationship is considered to be the most aesthetic<sup>1, 12</sup>.

Regarding the buccal corridor space, Ker et al.<sup>13</sup> established a maximum tolerable buccal corridor width of 1.6, an ideal width of 1.16, and a minimum tolerable width at 0.58. Our mean width of 0.59 falls just above the minimum tolerable threshold, suggesting that while some individuals may have a less ideal buccal corridor, their width is still within acceptable limits. According to Machado<sup>4</sup>, a wider buccal corridor is considered to be unattractive, while an intermediate width is more aesthetic. Our findings support this view, indicating that the participants' buccal corridors, while close to the minimum threshold, do not significantly detract from their smile aesthetics.

Ackerman and Ackerman<sup>14, 15</sup> originally developed the smile index, which is a crucial indicator of an attractive smile. An attractive smile is frequently considered to have a smile index of  $> 5.0$ . Wang et al.<sup>3</sup> showed a smile index of 6.02 for girls and 6.31 for boys in China. The smile index for Japanese women was ranged from 5.37 to 7.0<sup>16</sup>. The mean smile index in our study was 5.85, which aligns with the attractive range identified in previous research. Hence, our participants' smiles generally fall within the aesthetically pleasing category.

Another crucial metric for assessing smile aesthetics is dynamic smile symmetry, which may be calculated using the method described by Wang et al.<sup>3</sup> A smile that is visually appealing usually has a symmetry value of approximately 1. Our Vietnamese participants had a dynamic smile symmetry of 1.01 with a standard deviation of 0.06, indicating that their smiles have a high degree of bilateral

uniformity. Our findings are consistent with the established standard for attractive smiles; in other words, our participants' smiles generally display a balance that is considered to be aesthetically pleasing.

We identified significant gender differences for the anterior smile line, the smile arc, the upper lip curvature, and the most posterior tooth displayed when smiling. Compared with the men, women displayed a parallel smile arc (57.5% vs 35.6%) and a straight upper lip curvature (42.5% vs 29.4%) more frequently. Regarding the anterior smile line, the women more often exhibited a high or very high smile line, while the men more often had a low or average smile line. There was a minor gender difference in the most posterior tooth displayed when smiling. Men and women predominantly displayed their second premolar (54.8% and 56.9%, respectively). However, men displayed their first molar (23.5%) more frequently than women (13.8%), suggesting a broader smile. Finally, there was slight difference in dynamic smile symmetry, which refers to the uniformity of movement of the smile. Women had a mean  $\pm$  SD of  $0.99 \pm 0.05$ , while men had a mean  $\pm$  SD of  $1.01 \pm 0.07$ . Although the difference was statistically significant, the clinical relevance of this difference may be minimal.

According to Pham,<sup>9</sup> attractive smiles are likely to have some features such as an average or a high anterior smile line, a parallel smile arc, an upward upper lip curvature, and display up to the second premolar. In the current research, 56% of the students had an average or a high anterior smile line, 43.1% of the participants had a parallel smile arc, 10.2% of the participants had an upward upper lip curvature, and 58.6% displayed the second premolar. When examining these criteria through the lens of gender differences, although fewer women than men had an average or a high anterior smile line (55.1% vs. 56.3%) and an upward upper lip curvature (9.6% vs. 10.5%), more women had a parallel smile arc (57.5% vs. 35.6%). Additionally, more women than men had a smile that extended to their second molar (56.9% vs. 54.8%), which closely aligns with the characteristics of attractive smiles.

We found significant gender differences in various smile-related oral characteristics. Regarding the four characteristics of an attractive smile, the women tended to have two of these features more frequently than the men: the smile arc and the most posterior

tooth displayed when smiling. On the other hand, the men tended to have more frequently the anterior smile line and upper lip curvature associated an attractive smile. These insights are crucial for dental professionals aiming to provide appropriate and effective aesthetic treatments.

Despite offering valuable insights, this study has several limitations. The sample consisted exclusively of university students aged 18-29 years, representing a relatively narrow and homogeneous age group. As a result, the generalizability of the findings to other age cohorts or Vietnamese populations with varying socio-demographic backgrounds may be limited. To enhance external validity and further substantiate the current findings, future research by our group

will involve broader age distributions and more diverse demographic profiles.

## CONCLUSION

We found significant gender differences in various smile-related oral characteristics. Regarding the four characteristics of an attractive smile, the women tended to have two of these features more frequently than the men: the smile arc and the most posterior tooth displayed when smiling. On the other hand, the men tended to have more frequently the anterior smile line and upper lip curvature associated an attractive smile. These insights are crucial for dental professionals aiming to provide appropriate and effective aesthetic treatments.

## CONFLICT OF INTERESTS

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

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# Hand file and manual rotary file behavior in curved canals: an ex vivo micro-CT study

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## ABSTRACT

Several instrumentations systems are available in the market and it is necessary to assess their behavior in curved root canals. **Aim:** To analyze the ability of two manual instrumentation techniques to center mesial root canals of mandibular molars, studied by computed microtomography (micro-CT). **Materials and Method:** Twenty mesial root canals of mandibular molars were matched based on similar morphological dimensions using micro-CT evaluation and divided in 2 groups (n=10): (1) Crown-down technique with Gates-Glidden drills and K-Flexofiles and (2) M manual NiTi rotary system. Changes in volume, surface area and canal transportation were compared using an unpaired t-test with a 5% significance level. **Results:** No significant differences were observed between groups regarding volume surface area after root canal preparation (p>0.05). Variation in the centroid differed between groups in the total canal length, and in the cervical and middle thirds, with better centralization for the M files (p<0.05). **Conclusions:** Both manual instrumentation techniques had similar volume and surface area variation. Both techniques left unprepared canal areas with similar values. M manual NiTi files caused minor canal transportation.

**Keywords:** molar teeth - root canal preparation - dental instruments - X ray microtomography

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## Comportamento de limas manuais e rotatórias acionadas à mão em canais curvos: um estudo ex vivo em micro-TC

## RESUMO

Vários sistemas de instrumentação estão disponíveis no mercado, e é necessário avaliar seu desempenho em canais radiculares curvos. **Objetivo:** Analisar, por microtomografia computadorizada (micro-CT), a capacidade de centralização dos canais radiculares mesiais de molares inferiores preparados com duas técnicas de instrumentação manual. **Materiais e Método:** Vinte canais radiculares mesiais de molares inferiores foram pareados com base em dimensões morfológicas semelhantes, utilizando avaliação por micro-CT e divididos em dois grupos (n=10): Técnica Crown-down com brocas Gates-Glidden e K-Flexofiles e sistema rotatório manual NiTi M. Alterações no volume, área de superfície e transporte do canal foram comparadas usando o teste t não pareado com um nível de significância de 5%. **Resultados:** Não foram observadas diferenças significativas entre os grupos em relação à área de superfície do volume após a preparação do canal radicular (p>0,05). A variação no centro de gravidade mostrou diferenças entre os grupos em todo o comprimento do canal e nos terços cervical e médio, com melhor centralização para os sistemas testados M (p<0,05). **Conclusões:** Ambas as técnicas de instrumentação manual apresentaram variação semelhante de volume e área de superfície. Ambas as técnicas deixaram áreas do canal não preparadas com valores semelhantes. Os sistemas manuais de NiTi M apresentaram menor transporte do canal.

**Palavras-chave:** dentes molares - preparo do canal radicular - instrumentos odontológicos - microtomografia de raios X

## INTRODUCTION

The mechanical objectives of root canal instrumentation are to prepare and shape the root canals without causing deviation or modifying their original path<sup>1</sup>, thereby improving the efficacy of irrigation and final obturation.

In recent years, advancements in NiTi metallurgical designs and heat treatments have significantly improved endodontic mechanized rotary instrumentation. For example, heat-treated NiTi files are more flexible and more resistant to cyclic fatigue. This technical evolution in modern endodontic instrumentation has led to several micro-CT studies comparing different methods of mechanized instrumentation<sup>2,3</sup>.

Some schools of dentistry still teach manual techniques using stainless-steel hand files for preparing root canals. Hand file instrumentation with K-files usually requires a longer learning curve and acquisition of clinical competence in technical procedures<sup>4</sup>. Moreover, manual techniques are usually time consuming and involve several steps and files to prepare the root canal<sup>5</sup>. Although K-files can be bent to negotiate canal curvatures, they do not achieve the desired instrumentation quality, especially in posterior teeth, since the rigidity of the instrument metal results in canal transportation<sup>4</sup>.

Mesial roots of mandibular molars usually have narrow canals, especially in the cervical third, in which their mesiodistal diameter is smaller than the buccolingual diameter<sup>6</sup>, and Gates-Glidden (GG) drills are frequently used to enlarge the cervical and middle third of the canals when manual techniques are used. Even though a previous study reported that GG drills were safe regarding dentin thickness removal<sup>7</sup>, their use often leads to procedural errors such canal deviation, strip perforations or excessive and irregular dentin removal<sup>5</sup>.

Although it has been well established that mechanized instrumentation with NiTi files has significant advantages over manual K-file techniques<sup>8,9</sup>, engine-driven instruments require costly investment in electric endodontic motors and NiTi files. Not all universities can afford this kind of equipment<sup>10</sup>, and students in some countries cannot afford them due to different socioeconomic profiles<sup>11</sup>. To overcome these limitations, some manufacturers created manual NiTi rotary files, which according to some studies, provide better results than K-file manual instrumentation techniques<sup>12,13</sup>. The M file (Easy

Bassi, Belo Horizonte, Brazil) is a manual NiTi rotary file system with controlled memory (CM) heat treatment, that consists of few instruments to prepare the canals in a crown-down technique. The system includes orifice shapers (15.10 and 15.08), instruments from 15.05 to 40.05 for most of the cases, and complementary finishing files 50.05, 60.05, and 70.05. NiTi heat-treated alloys such as CM have greater flexibility and higher resistance to cyclic fatigue compared to conventional NiTi instruments. Another advantage of this treatment is the controlled shape memory, which is the ability of an instrument to stay deformed after insertion into the curved canal, returning to their original shape only after heat is applied, thereby maintaining the canal centered curvatures, and decreasing the risk of ledging, transportation or perforation<sup>14</sup>.

X-ray computed microtomography (micro-CT) imaging has been widely used for quantitative and qualitative morphologic 2D and 3D analyses on the shaping ability of endodontic instruments and possible alterations in the original root canal path<sup>2,6</sup>. The aim of this study was therefore to use micro-CT technology to assess the differences between systems by comparing the shaping ability of manual instrumentation using a crown-down technique with Gates Glidden and K-files versus manual rotary instrumentation with M files in the mesial canals of mandibular molars. The null hypothesis was that there is no difference in shaping and centering ability between these two instrumentation techniques.

## MATERIALS AND METHOD

### Sample selection and initial scanning

This research was approved by the Londrina State University ethics committee (No, 93558018.8.0000.5231). The teeth used in this study were extracted for orthodontic or periodontal reasons and selected based on specific criteria, including similar morphological dimensions and moderate mesial root curvature (10° to 20° according to Schneider's classification)<sup>15</sup>. Twenty mandibular extracted first molars with Vertucci type IV canals (two independent canals in the mesial root) were selected from a pool of 422 stored micro-CT scanned teeth collection using the CTan software v1.14.4 (Bruker-microCT, Kontich, Belgium). The CTan software was also used to measure the total root length from tip of the root to the cemento-enamel

junction of each root, giving a median root length of 10.5 mm.

The endodontic access cavity was prepared with diamond burs, and prior to the initial micro-CT scan, the buccal side of each molar was marked at the cemento-enamel junction (CEJ) with a round diamond bur to facilitate differentiation of the MV and ML canals in the 3D micro-CT image processing. All teeth were scanned in a custom attachment using a Skyscan 1173 (Bruker-microCT, Kontich, Belgium) device. The scanning parameters were 80 kV, 90 mA, pixel size 12.1  $\mu\text{m}$ , 360° around the vertical axis, and rotation step 1.0, frame average (3) using a 0.5-mm-thick aluminum filter. Images of the mesial roots were reconstructed using the NRecon v.1.6.9 software (Bruker-microCT) with ring artifact correction 5, beam hardening correction 51%, and smoothing 5, to create axial and transverse slices of the internal structure. Initial analysis of volume and surface area were recorded using the CTAn v.1.14.4, (Bruker-microCT), and data homogeneity was confirmed ( $p > 0.05$ ).

### **Root canal instrumentation and final micro-CT scan**

Both mesial root canals were explored under magnification using a size 10 K-file (Dentsply-Maillefer Baillagues, Switzerland) until the instrument tip was visible at the apical foramen. Then, the working length (WL) was set 1.0 mm shorter than the apical foramen. To control variables related to anatomy of the mesial root system, both instruments were used in the same root although alternating the mesial canals from root to root<sup>6</sup>. Thus, 20 root canals were included per group. Each set of instruments was used to prepare 2 root canals.

### **Gates Glidden and K-Flexofile instrumentation group**

Instrumentation with K-Flexofiles was performed using a crown-down technique<sup>16</sup>. Initially, canals were explored with #10 and #15 K-files (Dentsply-Maillefer). Then, the cervical third was pre-enlarged using #40 and #35 K-flexofiles (Dentsply-Maillefer). Subsequently, #2 and #1 Gates-Glidden drills (Dentsply-Maillefer) were used to flare the straight part of the canal. The movement performed with the GG drill was slight apical pressure and ups and downs with only one penetration with each drill. Manual files were used with the balanced

force technique<sup>17</sup> until the full WL was achieved. After instrumentation, each file was removed from the canal and cleaned with moist gauze to remove the debris. Apical enlargement was completed at 35.02 apical diameter, and a step-back technique was performed with flexofiles #40.02 K (1 mm short of WL), #45.02 (2 mm short of WL), and #50.02 (3 mm short of WL), confirming canal patency with the master apical file. All canals were irrigated with 1 mL of 2.5% NaOCl (CloroRio, São José do Rio Preto, Brazil) using a disposable syringe and 27-G NaviTip needles (Ultradent Products, Inc., South Jordan, UT, USA) after each file or GG bur insertion. The total irrigation volume was 15 mL per canal.

### **M instrumentation group**

For instrumentation with M (Easy Bassi), an initial canal exploration was performed with #10 and #15 K-flexofiles, and the cervical portion of the root canal was enlarged with #15.05 and 15.08 M files. The instrument was inserted with clockwise (CW) and counterclockwise (CCW) movement using light apical pressure until no resistance was encountered. Then, rotation movements were performed to cut dentin. The file was removed from the canal and cleaned with moist gauze to remove the debris. A 25.05 file was used in the same manner until 2/3 of the canal length was reached. The apical portion of the canal was instrumented with a 15.05 M file and subsequently finished until the 35.05 files using the same movements described above. After instrumentation of each third, the canal was irrigated with 5 mL of 2.5% NaOCl using a disposable syringe and 27-G NaviTip needles with 15 mL of irrigation volume per canal.

Once instrumentation was complete in both groups, final irrigation with 5 mL of 2.5% NaOCl and 17% ethylenediaminetetraacetic acid (EDTA) (Biodinamica, Ibiporã, Brazil) was applied to remove the smear layer. The EDTA was removed with saline solution and the canals were dried with paper points (Dentsply-Maillefer).

### **Volume, surface area and unprepared area analysis**

Reconstructed images acquired after instrumentation were geometrically co-registered with the preoperative data sets using the 3D Slicer 4.4.0 software (<http://www.slicer.org>) with a custom combination of a rigid registration module based on

image intensity similarities with accuracy greater than 1 voxel. All micro-CT analyses were performed by authors blinded to the instrumentation system groups. The analysis included the binarization of the root canals and measurement of the total canal volume ( $\text{mm}^3$ ) and surface area ( $\text{mm}^2$ ) using the CTAn v.1.14.4 software (Bruker). The unprepared surface of the root canal length was analyzed using the ImageJ 1.50 d software (National Institutes of Health, Bethesda, MD) by calculating the number of static voxels. All values were calculated by subtracting the scores for the treated canals from those recorded for their untreated counterparts and then converted into percentages.

The CTVol v.2.3.1 software (Bruker micro-CT) was used to define a color-coded standard for root canal models (green for preoperative canal, red for Manual K-flexofile instrumentation, and blue for M files postoperative canal surfaces), enabling a qualitative comparison of the superimposed root canal models before and after preparation.

### Canal transportation

Canal transportation was assessed as specified in a previous study<sup>8</sup>. The centers of gravity were calculated for each slice and connected along the z-axis with a fitted line in a total of 14.040 Manual K-flexofiles and 15.866 M Files cross sections using XLSTAT-3DPlot for Windows (Addinsoft,

New York, NY). Mean transportation (in mm) was calculated by comparing the centers of gravity before and after preparation for the coronal, middle and apical canal segments. Representative measurements were also graphically presented in diagrams (Figs. 1 and 2).

### Statistical analysis

Data distribution was checked for normality using the Shapiro–Wilk test. An unpaired T-test was used for group comparison of volume, surface area, unprepared surface areas and canal transportation before and after preparation. For intergroup comparison, a paired t-test was used. All values were analyzed using Prism 7.0 (GraphPad Software, Inc., La Jolla, CA, USA) and expressed as mean and standard deviation, and the median, minimum and maximum values were presented. The significance level was set at 5%.

## RESULTS

### Volume, surface area and unprepared areas

Volume, surface area and unprepared canal surface areas data before and after preparation are shown in Table 1. No statistical differences were found between groups in the initial and final canal volume and surface area changes ( $p>0.05$ ). Nonetheless, these values significantly increased after preparation with both systems in the intragroup comparison

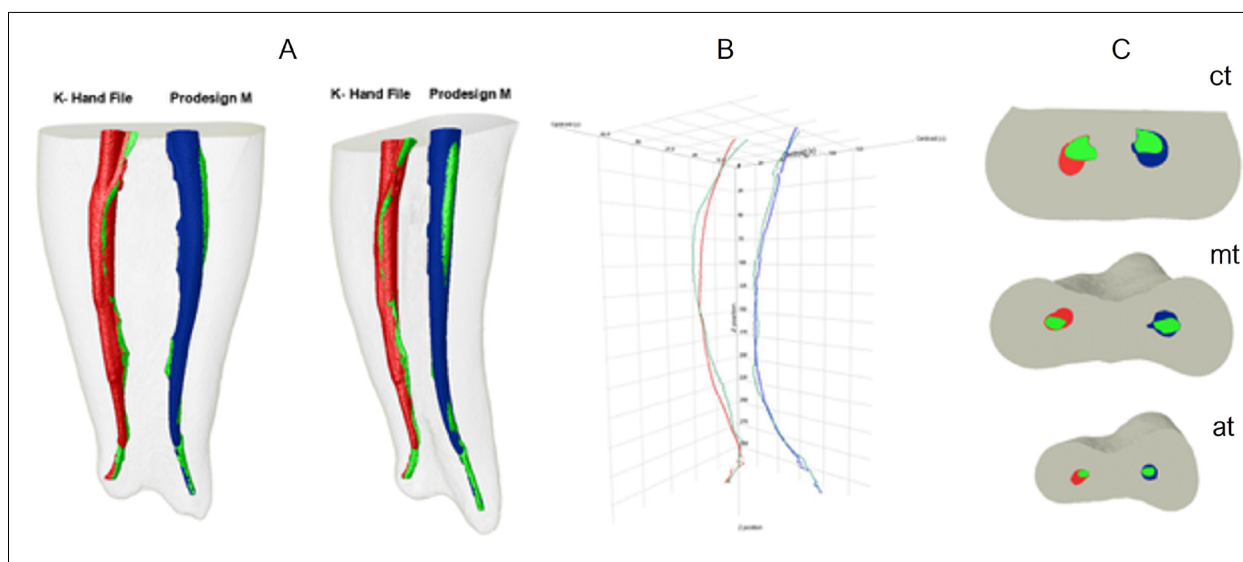


Fig. 1: A) Representative three-dimensional images of micro-CT scan taken before (green) and after preparation using either K-Flexofiles (red) or M files (blue) instruments. Superimposed views show unprepared areas in green. (B) Graph shows the centroid variation before (green) and after: K-Flexofiles (red) and M files (blue) canal preparation. (C) Cross-sectional views of the canal changes before and after coronal (ct), middle (mt), and apical (at) canal thirds.

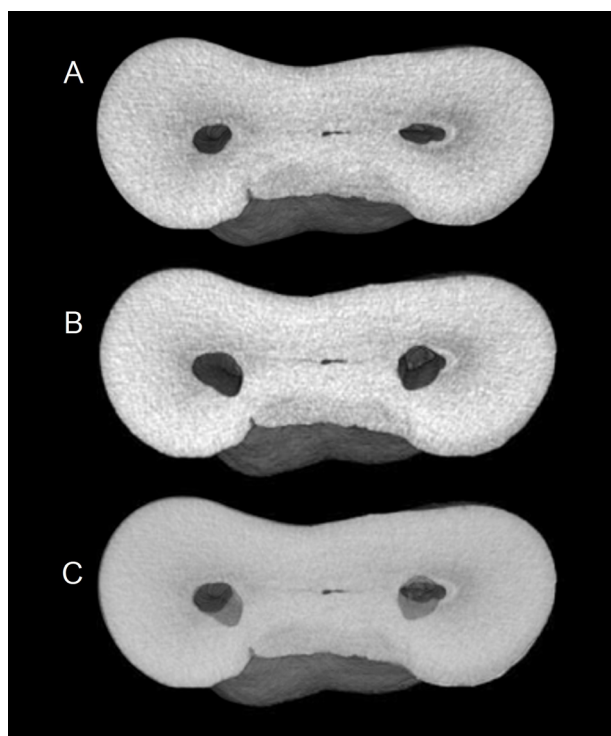


Fig. 2: Representative images of the dentin removal with each instrument towards the furcation area, located at 3 mm from the canal orifice. Left canal images were instrumented with GG/K-flexofiles and right canal images were instrumented with M files rotary Niti files. A) preoperative images, B) postoperative images, and C) superimposed images of A and B.

( $p < 0.05$ ) (Table 1). Regarding the unprepared surface area of the canals, K-flexofile instrumentation yields around 22 % while M files yield 18 % of unprepared areas, without significant differences between systems ( $p > 0.05$ ).

## Canal Transportation

Canal transportation data is shown in Table 2. Variation in the centroid differed significantly between groups in the total canal length, and in the comparison of the cervical and middle thirds ( $p < 0.05$ ). At the apical level, the centroid shift was higher for K-file (0.69 mm) than M file (0.51 mm) instrumentation, though without statistical differences ( $p > 0.05$ ). The mean scores for total canal length were 0.88 mm for K-flexofiles and 0.58 mm for M files. The comparison of the cervical, middle and apical thirds within the same groups showed significant differences between the apical level vs. both cervical and middle thirds in the K-flexofile group ( $p < 0.05$ ), but no significant difference in the M-file group ( $p < 0.05$ ).

## DISCUSSION

Mesial roots of mandibular molars were used to compare two manual techniques. To reduce biases, only one person with experience in both manual techniques prepared all the root canals. Regarding the root canal anatomical variations, canals with similar curvatures, pre-operative volumes and surface areas were selected. The instrumentation of the mesiobuccal and mesiolingual canals were alternated from specimen to specimen, enabling comparison of the two techniques under similar anatomical conditions<sup>18</sup>.

According to the micro-CT morphological analyses of volume and surface area, there was no statistical difference between groups ( $p > 0.05$ ). In the intragroup comparison, canal volumes and surface

**Table 1. Volume and surface area of mandibular molar mesial root canals before and after instrumentation with both systems.**

| Instrument system | Volume (mm <sup>3</sup> ) | Mean $\pm$ SD     | Median (range)         | Surface Area (mm <sup>2</sup> ) | Mean $\pm$ SD     | Median (range)        | Unprepared surface area (%) | Median (range)      |
|-------------------|---------------------------|-------------------|------------------------|---------------------------------|-------------------|-----------------------|-----------------------------|---------------------|
| K-Flexofile       | Initial                   | 1.80 $\pm$ 0.69   | 1.77 (0.77 - 3.65)     | Initial                         | 20.05 $\pm$ 4.42  | 20.75 (10.87 - 26.71) |                             |                     |
|                   | Final                     | 4.08 $\pm$ 0.59   | 4.12 (3.02 $\pm$ 5.06) | Final                           | 27.28 $\pm$ 3.88  | 27.71 (20.81 - 36.22) | 22.32 $\pm$ 11.3            | 17.93 (8.57 - 44.3) |
|                   | % increase                | 55.92 $\pm$ 14.90 | 58.81 (25.81 - 81.02)  | % increase                      | 25.95 $\pm$ 15.90 | 26.16 (4.61 - 55.25)  |                             |                     |
| M files           | Initial                   | 1.90 $\pm$ 0.81   | 1.40 (0.7 - 4.21)      | Initial                         | 19.83 $\pm$ 4.92  | 20.98 (11.99 - 28.02) |                             |                     |
|                   | Final                     | 3.69 $\pm$ 1.04   | 4.18 (2.49 - 5.77)     | Final                           | 25.58 $\pm$ 4.01  | 25.43 (19.39 - 32.19) | 18.58 $\pm$ 10.15           | 15.81 (6.92 - 8.43) |
|                   | % increase                | 48.42 $\pm$ 15.00 | 44.70 (26.66 - 75.91)  | % increase                      | 22.75 $\pm$ 13.53 | 18.26 (7.50 - 50.14)  |                             |                     |

**Table 2. Centroid shift (mm) in root canals after preparation with two manual instrument systems.**

| Instrument system   | Level              | Mean $\pm$ SD                 | Median | Range       | 25 -75 percentile |
|---------------------|--------------------|-------------------------------|--------|-------------|-------------------|
| <b>K-flexofiles</b> | Cervical           | 1.17 $\pm$ 0.42 <sup>aA</sup> | 1.13   | 0.46 - 1.84 | 0.77 - 1.48       |
|                     | Middle             | 1.09 $\pm$ 0.58 <sup>aA</sup> | 1.12   | 0.10 - 1.91 | 0.65 - 1.58       |
|                     | Apical             | 0.69 $\pm$ 0.43 <sup>aB</sup> | 0.74   | 0.04 - 1.58 | 0.34 - 1.01       |
|                     | Total canal length | 0.88 $\pm$ 0.51 <sup>a</sup>  | 0.94   | 0.01 - 1.96 | 0.49 - 1.10       |
| <b>M files</b>      | Cervical           | 0.51 $\pm$ 0.31 <sup>bA</sup> | 0.52   | 0.01 - 1.10 | 0.21 - 0.73       |
|                     | Middle             | 0.68 $\pm$ 0.56 <sup>bA</sup> | 0.62   | 0.02 - 2.06 | 0.19 - 0.92       |
|                     | Apical             | 0.51 $\pm$ 0.33 <sup>aA</sup> | 0.46   | 0.09 - 1.47 | 0.26 - 0.64       |
|                     | Total canal length | 0.58 $\pm$ 0.34 <sup>b</sup>  | 0.51   | 0.00 - 1-10 | 0.34 - 0.91       |

Different lowercase letters in the mean column indicate significant differences between the Manual K-flex and M instruments. Different uppercase letters indicate significant differences between different thirds in the same group.

area increased significantly ( $p > 0.05$ ). Nevertheless, 22 % (K-flexofile) and 18 % (M) of unprepared canal surface areas were observed after root canal instrumentation. Although canal surface area or volume increase could improve irrigation, from the mechanical standpoint, it does not mean that all canal walls will be completely cleaned<sup>19</sup>. Tissue remnants, debris and unaltered bacteria biofilms can remain harbored in canal irregularities and attached to the unprepared canal walls<sup>20</sup>. Thus, chemical irrigation solutions alone cannot be expected to clean these areas, so supplementary instrumentation techniques<sup>21</sup> and final irrigation protocols such as passive ultrasonic irrigation (e.g., Xpendo Finisher, among others), are recommended<sup>22</sup>.

When canal curvatures are present, there is a tendency for all preparation techniques to divert the prepared canal away from the original axis<sup>23</sup>. However, in this study, only the apical 2 mm were measured in linear mesiodistal direction using CBCT images. In our study, canal transportation in the apical portion of the canals did not differ significantly between groups. Our method using a more accurate imaging device (Micro-Ct)<sup>2</sup> was based on calculation of the centers of gravity for each slice connected along the z-axis with a fitted line. Thus, more cross-sections were analyzed, which might provide more accurate measurements. Another possible explanation for the minimal apical transportation in our study could be that, by first enlarging the coronal and middle portion of the root, both files (pre-curved K and NiTi rotary hand files) instrumented the apical portion of the canal with low deviation values. The final apical enlargement up to a size 35 was chosen based on a previous study which showed significantly higher

root canal disinfection after large apical diameters<sup>24</sup>. In mesial roots of mandibular molars, the pericervical dentine must be removed carefully, avoiding excess removal and deviation towards the danger zone<sup>25</sup>, which can weaken the root. A previous study reported that the risk of root fracture increases when the canal width is larger than 40% of the root width<sup>16,17,25</sup>. Although analysis the remaining dentin thickness towards the inner portion was not part of the aim of this study, the crown-down technique with GG drills and K-files significantly deviated the original canal centroid, rejecting our null hypothesis (Table 3).

Our results showed that deviation in the coronal portion with GG drills was more than 0.50 mm compared to the M-files, especially towards the furcation area (Figs.1 and 2, Table 2). Gates-Glidden drills were used to instrument the cervical third as orifice openers in the K-file group. However, when M-files were used, a 15.08 orifice shaper, which is more conservative, was used for pre-flaring the cervical third. In our study, GG drills caused more deviation than M-files. These differences might be explained by instrument design, rigidity, rpm needed (20,000) to increase cutting ability, which might produce a fulcrum effect at the orifice level which would move the Gates Glidden blade of the bur towards the furcal aspect of the canal wall<sup>25</sup>.

## CONCLUSIONS

In this *ex vivo* study, both manual instrumentation techniques produced similar enlargement of the root canal in relation to volume and surface area. Neither technique achieved 100% of unprepared canal areas, presenting similar values. The M manual

**Table 3. Centroid shift (mm) in the root canals after preparation with two manual instrument systems.**

| Instrument system   | Level              | Mean $\pm$ SD                 | Median | Range       | 25 -75 percentile |
|---------------------|--------------------|-------------------------------|--------|-------------|-------------------|
| <b>K-flexofiles</b> | Cervical           | 1.17 $\pm$ 0.42 <sup>aA</sup> | 1.13   | 0.46 - 1.84 | 0.77 - 1.48       |
|                     | Middle             | 1.09 $\pm$ 0.58 <sup>aA</sup> | 1.12   | 0.10 - 1.91 | 0.65 - 1.58       |
|                     | Apical             | 0.69 $\pm$ 0.43 <sup>aB</sup> | 0.74   | 0.04 - 1.58 | 0.34 - 1.01       |
|                     | Total canal length | 0.88 $\pm$ 0.51 <sup>a</sup>  | 0.94   | 0.01 - 1.96 | 0.49 - 1.10       |
| <b>M files</b>      | Cervical           | 0.51 $\pm$ 0.31 <sup>bA</sup> | 0.52   | 0.01 - 1.10 | 0.21 - 0.73       |
|                     | Middle             | 0.68 $\pm$ 0.56 <sup>bA</sup> | 0.62   | 0.02 - 2.06 | 0.19 - 0.92       |
|                     | Apical             | 0.51 $\pm$ 0.33 <sup>aA</sup> | 0.46   | 0.09 - 1.47 | 0.26 - 0.64       |
|                     | Total canal length | 0.58 $\pm$ 0.34 <sup>b</sup>  | 0.51   | 0.00 - 1.10 | 0.34 - 0.91       |

Different lowercase letters in the mean column represent significant differences between the Manual K-flex and M files instruments. Different uppercase letters represent significant differences between different thirds in the same group.

NiTi system maintained the original canal path with significantly less canal transportation than did the

K-Flexofiles crown-down technique.

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# Level of agreement among dental students and general dentists on the diagnosis of periodontitis using the new classification of periodontal diseases

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## ABSTRACT

The new classification system of periodontal diseases, introduced in 2017, aims to provide more precise diagnostic criteria for identifying and classifying periodontitis. It defines periodontal disease based on staging, grading and extent, facilitating a more accurate approach for general dentists and periodontists. **Aim:** The aim of this study was to establish the inter-observer agreement and diagnostic accuracy among three groups with different levels of education and experience in dentistry in assigning stages, grades and extent of periodontitis using the 2017 AAP-EFP classification system. **Materials and Method:** The study involved 32 participants divided into three groups: Group 1-dental students (n=12), Group 2-dentists with up to 3 years' experience (n=10), and Group 3-general dentists with more than 4 years' clinical experience (n=10). Twelve periodontitis cases were evaluated by all participants, who classified each case by stage, grade and extent. The evaluations were compared with the gold standard provided by a panel of expert periodontists. Statistical analysis, including Fleiss' kappa and chi-square tests, was performed to assess agreement and diagnostic accuracy. **Results:** Significant differences were observed in stage classification, with students and recent graduates more likely to underestimate the severity of the disease, while experienced dentists tended to overestimate it. No significant differences were found in grade and extent classifications. Diagnostic accuracy was comparable across all groups, but students showed slightly higher agreement with the gold standard compared to other groups. **Conclusions:** The 2017 AAP-EFP classification system can be applied by both general dentists and dental students, but further clinical practice is needed to improve diagnostic accuracy. Misclassification tendencies, such as over- or underestimation of disease severity, highlight the need for additional training and familiarity with the system.

**Keywords:** periodontitis - diagnosis - sensitivity and specificity - dental students - dental education - reproducibility of results

## Nivel de concordancia en el diagnóstico de periodontitis utilizando la nueva clasificación de las enfermedades periodontales entre estudiantes de odontología y odontólogos generales

### RESUMEN

La nueva clasificación de las enfermedades periodontales, introducida en 2017, ofrece criterios diagnósticos más precisos para identificar y clasificar la periodontitis. Este sistema clasifica la enfermedad periodontal en función de un estadio, grado y extensión, permitiendo un abordaje diagnóstico más objetivo tanto para odontólogos generales como para periodoncistas. **Objetivo:** Establecer el grado de concordancia interobservador y la precisión diagnóstica en la asignación de estadios, grados y extensión de la periodontitis utilizando la clasificación AAP-EFP 2017, entre tres grupos con distintos niveles de formación y experiencia en odontología. **Materiales y Método:** Se reclutaron 32 participantes, distribuidos en tres grupos: Grupo 1-estudiantes de odontología (n=12), Grupo 2-odontólogos con hasta 3 años de experiencia clínica (n=10) y Grupo 3-odontólogos generales con más de 4 años de práctica clínica (n=10). Los participantes evaluaron 12 casos clínicos de periodontitis, asignando estadio, grado y extensión a cada uno. Estas evaluaciones fueron comparadas con el estándar de referencia establecidos por un panel de expertos en periodoncia. Se utilizó el coeficiente kappa de Fleiss y la prueba de chi-cuadrado para determinar la concordancia y precisión diagnóstica. **Resultados:** Se observaron diferencias estadísticamente significativas en la clasificación del estadio de la periodontitis, con una tendencia a la subestimación de la severidad de la enfermedad en los estudiantes y recién graduados, frente a una tendencia a la sobreestimación en los odontólogos con mayor experiencia clínica. No se encontraron diferencias significativas en la clasificación del grado y la extensión de la enfermedad entre los grupos. La precisión diagnóstica fue comparable entre todos los grupos, observándose una mayor concordancia del grupo de los estudiantes con el gold standard. **Conclusiones:** El sistema de clasificación de 2017 de la AAP-EFP puede ser utilizado tanto por odontólogos generales como por estudiantes, sin embargo se requiere mayor práctica clínica para mejorar la precisión diagnóstica. Las tendencias a clasificar erróneamente la severidad de la enfermedad, ya sea por sobreestimación o subestimación, subrayan la necesidad de entrenamiento adicional y familiarización con el sistema para mejorar los resultados clínicos y la planificación de los tratamientos.

**Palabras clave:** periodontitis - diagnóstico - sensibilidad y especificidad - estudiantes de odontología - educación en odontología - reproducibilidad de los resultados

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## INTRODUCTION

At the 2017 World Workshop on the Classification of Periodontal and Peri-Implant Diseases and Conditions<sup>1</sup>, a new system for classifying periodontal diseases was established. This system defined a periodontitis case and introduced the assignment of stages, grades and extent to describe the severity, progression rate, treatment response and number of teeth involved in the diagnosis. This new classification system aims to provide a more precise diagnosis and is intended for implementation in clinical practice by both specialists and general practitioners. It is designed to be dynamic, enabling updates over time.

The need for a new classification was based on the deficiencies of the previous system (1999), which had imprecise criteria for determining the diagnosis, severity and complexities of periodontitis. The 1999 American Academy of Periodontology (AAP) Workshop established a classification system based on the scientific evidence available at the time, while the European Federation of Periodontology (EFP) did the same a year later<sup>2</sup>.

The 2017 Workshop was a joint effort by the AAP and the EFP, with contributions from various working groups that analyzed the current evidence and reached a consensus in their publication<sup>3,4</sup>.

A significant difference in this new system was the establishment of a clear definition of a periodontitis case, characterized by interproximal attachment loss in at least two non-adjacent teeth<sup>5,6</sup>.

Stages determine the severity of the disease, treatment complexity and percentage of affected teeth, while the grade provides information on the biological characteristics of the disease, including its progression rate, risk of further progression, and potential treatment response.

Stage is determined based on a complete series of radiographs of both arches, a full periodontal chart, and a detailed patient history. Grade is defined according to clinical periodontal records (periodontal chart) and full radiographs of both arches, which may be adjusted based on the presence of modifying risk factors such as smoking and diabetes<sup>7</sup>. Extent is determined by the percentage of affected teeth.

The definition of stages, grades and extent in cases of periodontitis using the new classification system requires a learning curve, not only for general practitioners but also for specialists in periodontics. Diagnostic accuracy is evaluated by comparing the

stage, grade and extent assignments between the examiner groups and an expert panel (gold standard). Identifying the level of diagnostic accuracy among study participants will provide valuable information to improve the implementation of this system across all groups of practitioners<sup>8</sup>. The dissemination within the dental community of this tool for making precise diagnoses will lead to appropriate treatment planning and follow-up for patients with periodontal disease<sup>9</sup>. Several studies have assessed the reliability and accuracy of the 2017 AAP-EFP classification system for periodontitis across different professional backgrounds and levels of expertise. Ravidà et al. analyzed the degree of agreement among international periodontal experts, and found notable variability even within this highly trained group<sup>8</sup>. Oh et al. reported significant discrepancies in periodontitis classification among dental practitioners with varying educational backgrounds, suggesting that training plays a critical role in diagnostic consistency<sup>10</sup>. Abrahamian et al. evaluated both intra- and inter-examiner reliability in applying the new classification, emphasizing the challenges of standardizing diagnosis across clinicians<sup>11</sup>. Similarly, Marini et al. compared the consistency and accuracy of staging and grading among periodontal specialists, general dentists and undergraduate students, revealing substantial differences between groups<sup>12</sup>.

To date, no study in Argentina has analyzed the diagnostic accuracy of periodontitis among dentists using the new classification system. The ability to distinguish between less severe (Stage I and II) and more severe cases (Stage III and IV) is crucial for both general practitioners and undergraduate trainees.

The aim of this research was to establish the inter-observer agreement and diagnostic accuracy in assigning stages, grades and extent of periodontitis using the 2017 AAP-EFP classification system among three groups with different levels of education and experience in dentistry.

## MATERIALS AND METHOD

### Study Design

This is an observational study on inter-observer agreement. The main objective was to evaluate the diagnostic accuracy and agreement level among dental students, general dentists with up to

3 years' clinical experience, and general dentists with more than 4 years' experience in classifying cases of periodontitis using the 2017 Classification of Periodontal Diseases. The evaluations were compared to those made by a panel of periodontal experts as the gold standard. The study was conducted according to the Guidelines for Reporting Reliability and Agreement Studies<sup>13</sup>.

### Population

The study included 32 examiners, divided into three groups:

- Group 1: dental students from 5<sup>th</sup> and 6<sup>th</sup> year at the Instituto Universitario Italiano de Rosario (IUNIR) (n= 12).
- Group 2: general dentists who recently graduated from IUNIR, with up to 3 years' clinical experience (n=10).
- Group 3: general dentists recruited from the teaching staff at IUNIR, with more than 4 years' clinical experience, without advanced training in periodontics (n=10).

All examiners signed informed consent prior to participation. Data collection was performed in April 2023.

### Inclusion Criteria

Group 1: Undergraduate dental students in their final years of study.

Group 2: General dentists with up to 3 years' clinical experience.

Group 3: General dentists with more than 4 years' clinical experience.

### Exclusion Criteria

Dentists with specialization or advanced postgraduate training in periodontics.

### Elimination Criteria

Examiners who did not complete all the required evaluations.

### Clinical Cases

Twelve representative cases of different stages and grades of periodontitis were selected from the patient database of the Periodontology Clinic at IUNIR. These cases were chosen by the Principal Investigator (FH) and a certified periodontics specialist. Cases of necrotizing periodontitis or

those with underlying systemic conditions were not included.

Each clinical case included the following information:

- a) Patient's age and gender.
- b) Relevant dental and medical history, including systemic diseases and risk factors such as diabetes (HbA1c values) and smoking.
- c) Intraoral photographs.
- d) Complete periodontal chart.
- e) Full series of radiographs.

### Expert Panel (Gold Standard)

The expert panel consisted of the Principal Investigator (FH) and another specialist in periodontics, who evaluated the 12 clinical cases using the 2017 New Classification. The consensus diagnosis for stage, grade and extent of each case was considered the gold standard for comparison to the assessments made by the examiners.

### Training Procedure

Prior to evaluating the cases, the examiners participated in a 3-hour training seminar on the 2017 New Classification of Periodontal Diseases. During the seminar, three clinical cases not included in the study were discussed, and relevant literature was provided. Examiners were able to clarify any questions regarding the classification before starting the formal evaluation of the 12 cases.

### Case Evaluation

Each examiner was granted access to a virtual classroom created for the project, where they were presented with the complete documentation of the 12 clinical cases. The examiners classified each case in terms of stage, grade, and extent using a standardized questionnaire based on the study by Oh et al., which included questions about their training, prior knowledge of the classification system, and its use in clinical practice<sup>10</sup>.

Overall agreement was defined as the global diagnostic consistency between each examiner group and the gold standard across the three classification domains—stage, grade and extent—considered collectively. This composite measure reflects the extent to which participants' overall diagnostic assessments matched those of the expert panel, providing a summary index of diagnostic accuracy beyond individual category agreement.

### Description of Periodontitis Case Characteristics

Twelve cases of periodontitis were evaluated, with patient ages ranging from 22 to 64 years (mean age: 42 years). Of the 12 cases, 4 were male patients and 8 were female. Five patients reported a family history of periodontal disease. Regarding smoking habits, 7 patients were non-smokers, 3 patients smoked fewer than 10 cigarettes per day, and 2 patients smoked more than 10 cigarettes per day. In terms of glycemic status, 10 patients were normoglycemic/non-diabetic, 1 patient was diabetic with HbA1c < 7%, and 1 patient was diabetic with HbA1c > 7%. One patient reported being a hepatitis C carrier, and 2 patients were HIV-positive.

### Statistical Analysis

Inter-observer agreement was evaluated using Fleiss' kappa statistics to measure the consistency in the classification of stage, grade and extent among the three groups of examiners.

Diagnostic accuracy was determined by comparing the examiners' classifications with the gold standard diagnosis, calculating the weighted quadratic kappa for each pairwise comparison. Frequencies and percentages of overall agreement were also calculated. A sub-analysis was performed to investigate the effect of variables such as stage, grade and modifying factors (smoking, diabetes) on diagnostic accuracy<sup>14</sup>. Differences in diagnostic accuracy among examiner groups were evaluated using the chi-square test, with a significance level of 0.05. Statistical analysis was conducted using IBM SPSS Statistics version 25.

### Ethical Considerations

This study was approved by the IUNIR Ethics and Bioethics Committee (CEB Res. 45/22). All participants signed informed consent before participating in the study. Patient confidentiality was safeguarded by removing all identifying information from the clinical cases presented to the examiners.

## RESULTS

### Stage Classification

Statistically significant differences between groups were observed in the stage assignment ( $p$ -value < 0.05), showing a general trend toward underestimation. Examiners tended to underestimate the stage of periodontitis more frequently than overestimate it (Fig. 1).

The Quadratic Weighted Kappa (QWK) values for stage classification showed moderate agreement across all examiner group comparisons:  $\kappa$  = 0.561 (Group 2 vs. Group 1),  $\kappa$  = 0.451 (Group 3 vs. Group 2), and  $\kappa$  = 0.376 (Group 3 vs. Group 1), with all  $p$ -values < 0.0001 (Table 1).

### Grade Classification

In the assessment of grade, no statistically significant differences were observed between the groups in terms of frequency distributions (Fig. 2). However, dentists with more than four years' experience (Group 3) tended to underestimate the disease grade more often, although the difference was not statistically significant.

QWK values indicated fair to moderate agreement:  $\kappa$  = 0.387 (Group 3 vs. Group 2),  $\kappa$  = 0.338 (Group

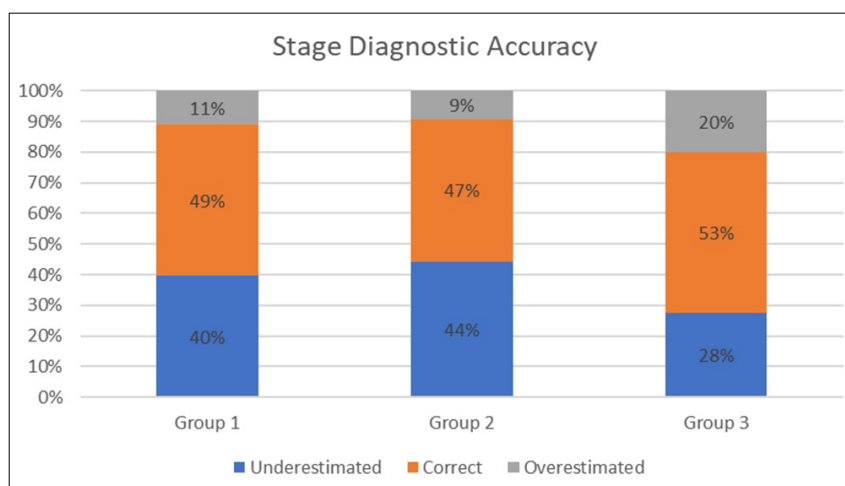


Fig. 1: Stage classification accuracy. Distribution of responses by group according to periodontitis stage classification ( $p$  < 0.05).

**Table 1. Fleiss' kappa Statistics**

| Periodontitis | Index   | Dentist vs. Student | Professor vs. Student | Professor vs. Dentist |
|---------------|---------|---------------------|-----------------------|-----------------------|
| Stage         | Kappa   | 0.2857              | -0.0693               | 0.0199                |
|               | p-value | 0.0016              | 0.4475                | 0.826                 |
| Grade         | Kappa   | 0.2688              | 0.0917                | 0.1657                |
|               | p-value | 0.0019              | 0.2936                | 0.066                 |
| Extent        | Kappa   | 0.4449              | 0.2953                | 0.25                  |
|               | p-value | 0.0001              | 0.0011                | 0.006                 |
| Overall       | Kappa   | 0.3219              | 0.1268                | 0.1795                |
|               | p-value | 0.0001              | 0.0152                | 0.0007                |

2 vs. Group 1), and  $\kappa = 0.313$  (Group 3 vs. Group 1), all statistically significant ( $p < 0.0001$ ).

### Extent Classification

Regarding the classification of disease extent (localized vs. generalized), no significant differences were found among examiner groups ( $p > 0.05$ ) (Fig. 3).

Agreement levels, as measured by QWK, showed moderate consistency across all group comparisons:  $\kappa = 0.552$  (Group 2 vs. Group 1),  $\kappa = 0.436$  (Group 3 vs. Group 1), and  $\kappa = 0.415$  (Group 3 vs. Group 2), with all p-values  $< 0.0001$ .

### Diagnostic Accuracy

Comparison of the examiners' diagnostic accuracy to the gold standard (expert panel) showed no statistically significant differences overall (p-value = 0.4967). However, the highest agreement with the gold standard was observed in the student group

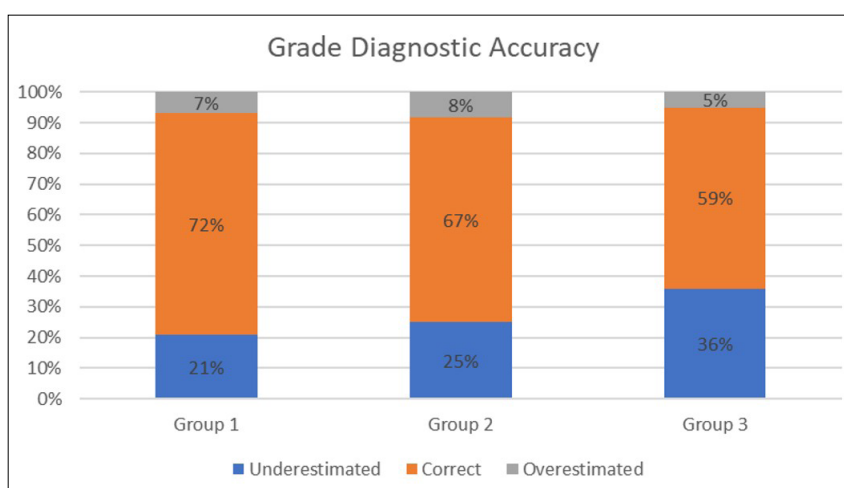


Fig. 2: Grade classification accuracy. Grade assignment by examiner group. No significant differences ( $p > 0.05$ ).

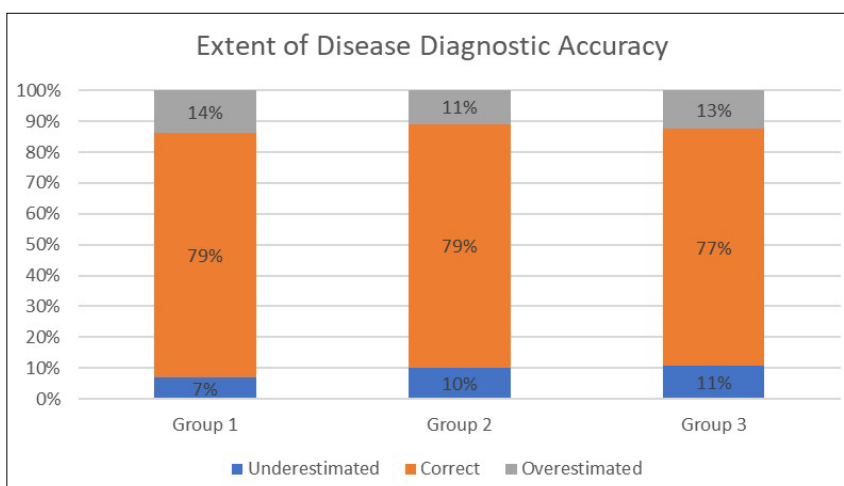


Fig. 3: Extent classification accuracy. Classification of disease extent among examiner groups ( $p > 0.05$ ).

(Group 1), followed by the dentists with less than 3 years' experience (Group 2). The dentists with more than 4 years' experience (Group 3) had the lowest concordance percentage (Fig. 4). In the classification of stage, students and less experienced dentists exhibited a greater tendency toward underestimation, while more experienced dentists showed a tendency to overestimate. Regarding grade, accuracy was similar for students and recently graduated dentists, while experienced dentists more frequently underestimated disease severity. For the extent of disease, no significant differences were observed among the examiner groups.

The overall QWK values indicated moderate agreement between groups, with the highest agreement observed between Group 2 and Group 1 ( $\kappa = 0.552$ ), and the lowest in the grading between Group 3 and Group 1 ( $\kappa = 0.383$ ). All kappa values were statistically significant ( $p < 0.0001$ ), reflecting systematic, though moderate, consistency in the application of the classification criteria across groups.

## DISCUSSION

The new classification system for periodontal diseases was introduced by the AAP and EFP in the 2017 Workshop and published in 2018. Its aim is to improve the identification and diagnosis of periodontitis and gingivitis by providing clear clinical and radiographic parameters and identifying significant risk factors. It can be used by both periodontists and general dentists<sup>1, 7</sup>.

This study was designed to analyze how dental

students and general dentists interpret and apply this new classification system. The examiners were divided into three groups: dental students, general dentists with up to three years' experience, and general dentists with more than four years' experience. Their assessments were compared against a gold standard established by an expert panel that diagnosed the clinical cases used in this study<sup>10</sup>.

The classification, which includes stage, grade and extent, enables more precise evaluation of the severity, progression and distribution of periodontal disease<sup>15</sup>. Although students had the advantage of recent training on the new classification, their diagnostic accuracy did not differ significantly from that of the other groups. Regarding stage evaluation, all groups agreed with the gold standard in approximately half the cases. However, students and newly graduated dentists tended to underestimate the stage, whereas dentists with greater clinical experience tended to overestimate it. This discrepancy may be due to a misinterpretation of clinical and radiographic parameters or a lack of familiarity with the new classification system. Marini et al. showed that overall, the consistency with the gold standard of general dentists was significantly lower than that of the other two groups<sup>12</sup>. In another study, Ravidà et al. reported agreement between raters and the gold-standard panel in terms of staging (76.6%), grading (82%) and extent (84.8%)<sup>8</sup>.

Periodontitis grade, which determines its progression rate, was estimated better by students and recently graduated dentists. In contrast, dentists

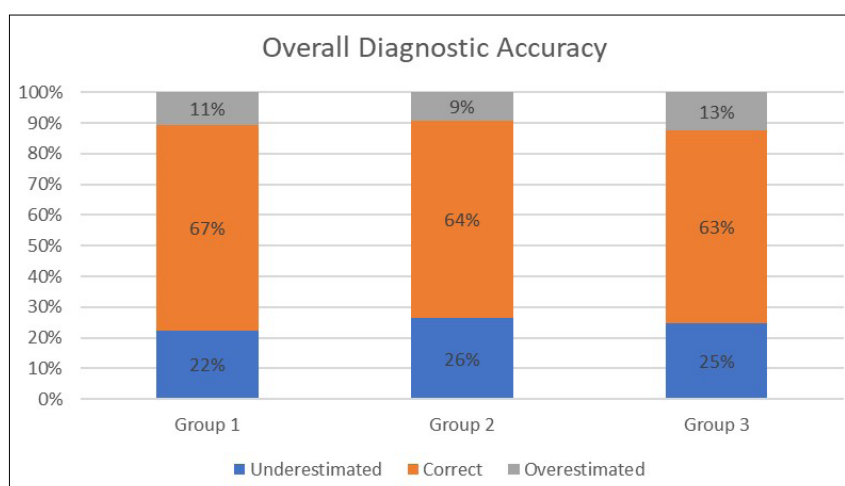


Fig. 4: Overall diagnostic agreement with the gold standard. Percentage of overall agreement by group across stage, grade and extent ( $p = 0.4967$ ).

with more than four years' experience had a higher error rate in their estimations. This discrepancy may be attributed to incorrect interpretation of indirect evidence, such as the relationship between radiographic bone loss and patient age. It is also possible that more experienced dentists did not fully consider modifying factors such as diabetes or smoking when estimating disease grade<sup>16, 17</sup>.

Winkler et al. concluded that smoking was a modifying factor in grade estimation<sup>17</sup>, which is consistent with the results of the current study. Oh et al. reported that the identification of risk factors for periodontal disease is also difficult, regardless of educational background, as indicated by the low level of accuracy and the lack of any significant difference in the recognition of risk factors<sup>10</sup>.

For periodontitis extent, all groups demonstrated similar levels of accuracy in classifying the disease as localized or generalized, making this the category with the least difficulty in interpretation.

The diagnosis of furcation lesions is crucial for determining periodontal disease staging. Our Exploring Furcation Involvement Diagnosis and Treatment Practices survey, published in 2023, identified a low percentage of furcation lesion detection among general dentists<sup>18</sup>. This finding is significant, as furcation involvement plays a key role in staging periodontitis within the new classification system, which is designed not only for specialists but also for general practitioners.

Finally, when analyzing the overall concordance between the groups and the gold standard across stage, grade and extent, no significant differences were observed. This suggests that, despite variations in interpretation, the overall diagnostic accuracy

is comparable between students and experienced dentists<sup>8, 11</sup>.

Previous studies, such as Marini et al., have shown that concordance with the gold standard is higher among students and specialists than among general dentists<sup>12</sup>. Similarly, Oh et al. reported that periodontists demonstrate superior diagnostic performance relative to general dentists<sup>10</sup>.

### Study Limitations

The main limitation of this study is the relatively short time since the implementation of the new classification system, which may explain the lack of familiarity among some participants, particularly more experienced dentists. Additionally, the use of clinical photographs instead of in-person evaluations may have limited the examiners' ability to make more precise diagnoses.

Future studies should include larger sample sizes and incorporate periodontal specialists as a separate analysis group to obtain more conclusive results.

### CONCLUSION

The findings of this study demonstrate that the 2017 AAP-EFP classification system for periodontitis can be applied with moderate consistency by dental students and general dentists with varying levels of clinical experience.

Although this study found an acceptable level of diagnostic accuracy, continued clinical practice and familiarity with the system will likely improve its interpretation and application. Incorporating structured educational interventions and case-based exercises may help reduce variability and improve diagnostic precision in clinical practice.

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### CONFLICT OF INTERESTS

The authors declare no conflicts of interest with respect to the research, authorship and/or publication of this article.

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# Oral health services in primary care: a study in southern Brazil

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## ABSTRACT

The evaluation of oral health services is an essential component of surveillance efforts aimed at improving health indicators and the quality of care. **Aim:** The aim of this cross-sectional study was to describe the characteristics of the oral healthcare work process in primary care settings in Porto Alegre, a major city in southern Brazil. **Materials and Method:** Data on patient profiles and process evaluation components were provided by the Porto Alegre Primary Healthcare Board. An electronic questionnaire was administered to dentists to assess the structure components. Data were classified into three modules: patient profile (sex and age), structure (human resources), and process (actions and procedures). Descriptive statistical analyses were conducted, including the calculation of absolute and relative frequencies. **Results:** In 2022, a total 414,682 dental appointments were recorded, the majority involving women and individuals aged 35–59 years. Most dentists were young women, primarily specialized in collective health. The process analysis highlighted a strong focus on preventive and health promotion actions, although fluoride applications, sealants, and cariostatic agents were underutilized. Composite resin restorations in permanent teeth were common. Dental emergencies accounted for 17% of cases, and 95% of surgical procedures consisted of extractions and sutures. Additionally, dentists performed integrative practices and other activities, such as rapid testing and vital sign measurements, particularly in response to the COVID-19 pandemic. **Conclusions:** The findings of this study are intended to inform strategies and service management aimed at improving oral health care in the primary care setting of Porto Alegre.

**Keywords:** primary healthcare - health services administration - oral health - dental care

## Serviços de saúde bucal na atenção primária: uma análise em uma grande cidade do sul do Brasil

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### RESUMO

A avaliação dos serviços de saúde bucal é um componente essencial dos esforços de vigilância que visam melhorar os indicadores de saúde e a qualidade do atendimento. **Objetivo:** Estudo transversal teve como objetivo descrever as características do processo de trabalho em saúde bucal na atenção primária em Porto Alegre, Rio Grande do Sul, Brasil. **Materiais e Método:** Os dados sobre o perfil de pacientes e de componentes da avaliação de processos foram fornecidos pela Diretoria de Atenção Primária à Saúde de Porto Alegre. Questionário eletrônico foi aplicado aos dentistas para avaliar o componente estrutura. Os dados foram categorizados em três módulos: perfil do paciente (sexo e idade), estrutura (recursos humanos) e processo (ações e procedimentos). Foram conduzidas análises estatísticas descritivas, incluindo o cálculo de frequências absolutas e relativas. **Resultados:** Em 2022, um total de 414.682 consultas odontológicas foram registradas, com a maioria envolvendo mulheres e indivíduos com idade entre 35 e 59 anos. A maioria dos dentistas eram mulheres jovens, e destacou-se a especialização em saúde coletiva. A análise de processo destacou um forte foco em ações preventivas e de promoção da saúde, embora aplicações de flúor, selantes e cariostáticos tenham sido subutilizados. Restaurações de resina composta em dentes permanentes eram comuns. Emergências odontológicas representaram 17% dos casos, e 95% dos procedimentos cirúrgicos consistiram em extrações e suturas. Além disso, os dentistas realizaram práticas integrativas e outras atividades, como testes rápidos e medições de sinais vitais, particularmente em resposta à pandemia de COVID-19. **Conclusões:** Os achados deste estudo pretendem destacar os pontos fortes e fracos da gestão de serviços visando melhorar os cuidados de saúde bucal no ambiente de atenção primária de Porto Alegre.

**Palavras-chave:** atenção primária à saúde - gestão de serviços de saúde - saúde bucal; assistência odontológica

## INTRODUCTION

Primary care is a fundamental pillar of the Brazilian national healthcare system, providing continuous care based on prevention, health promotion, treatment and rehabilitation<sup>1</sup>. Primary care serves as the gateway to Brazil's public healthcare system, as guaranteed by the Federal Constitution. It is the world's only universal healthcare system that provides free care to over 200 million individuals.

Despite the acknowledged importance of oral health for quality of life, Brazil's history in this field has been characterized by limited access and inadequate service provision. In response to the need for improved epidemiological indicators, the National Oral Health Policy was reorganized to expand access and enhance the quality of primary care, aiming to improve the population's oral health status<sup>2</sup>.

Epidemiological data provide the basis for designing studies that involve collecting and analyzing health information. Their primary purpose of such studies is to inform decision-making in health programs and services, guiding the planning and implementation of actions, while enabling the establishment of targets to improve overall performance<sup>3</sup>. The model proposed by Donabedian et al.<sup>4</sup> serves as a reference for evaluating the quality of healthcare services through three key components: structure, processes and outcomes. It is assumed that adequate structural conditions facilitate effective processes, which in turn lead to favorable health outcomes<sup>3-5</sup>.

The Brazilian Ministry of Health establishes management agreements with state and municipal health departments to guide the evaluation and monitoring of primary care within the public healthcare system. Viana et al.<sup>6</sup> examined the structure and process components of primary care in 168 municipalities in the state of Pernambuco, reporting higher average indicators of dental appointment records in contexts with better-organized work processes. Mendes et al.<sup>7</sup> investigated the relationship between dentists' professional profiles and the performance of oral health teams, concluding that continuing education and postgraduate qualifications positively influence the quality of dental services provided to the population. Similarly, Bueno et al.<sup>8</sup> analyzed the association between clinical productivity and dentists' academic backgrounds, finding that 60% of dentists had additional training and that specialists in public health demonstrated higher productivity

compared to those without a specialization or those specializing in other fields.

Studies evaluating oral health in primary care are limited in Brazil. Assessing practices and programs requires well-organized information systems as a key part of health surveillance. In this context, evaluating oral health interventions in primary care is crucial for strengthening programs and ensuring proper resource allocation to public healthcare services. The aim of this study was therefore to describe the patient profile and assess care quality using structural indicators (dentist profiles) and process indicators (actions and procedures) in primary care units in Porto Alegre, southern Brazil.

## MATERIALS AND METHOD

### Ethical considerations

This study was approved by the Research Ethics Committee of the Porto Alegre Municipal Health Department (certificate number: 6.105.210). The data are accessible to health administrators in the city of Porto Alegre, state of Rio Grande do Sul, Brazil. Participant confidentiality was ensured throughout the study.

### Study design

This was an observational, cross-sectional study with a descriptive component.

### Study setting

Porto Alegre, with a population of 1,332,570 residents and a Municipal Human Development Index (MHDI) of 0.8059, faces significant health challenges<sup>9</sup>. According to data from the city's Primary Care Policy, 27% of the population is at high risk of health vulnerability, while 80% of residents are registered at primary care units. The healthcare services are decentralized, organized into 17 health districts based on geographic boundaries<sup>10</sup>. As of 2022, the oral health primary care network in Porto Alegre consisted of 247 dentists distributed across 104 primary care units.

### Data collection

Data for the patient profile component and the evaluation of process indicators were provided by the Porto Alegre Primary Care Board, with a reference period from January to December 2022. To evaluate structure, data were collected using an electronic

questionnaire created in Qualtrics software, which was distributed to dentists via WhatsApp. The reference period for this data collection was August to October 2022.

Data collected for patient profiles included sex (male or female) and age group, which was subsequently classified. For dentist profiles, data collected included sex (male or female), age (in years), experience in primary care (years), academic background (years since graduation), academic title (*lato sensu and stricto sensu*), and specialty (according to the Federal Council of Dentistry). Clinical oral health procedures included in the list of services offered in primary care in Porto Alegre were grouped into three categories: type of care, active search for patients, and dental actions and procedures<sup>11</sup>. The data were entered into Microsoft Excel spreadsheets and organized into three modules: patient profile, structure evaluation and process evaluation, following the framework proposed by Donabedian et al.<sup>4</sup>.

### Data analysis

Descriptive statistics were performed, including the calculation of absolute and relative frequencies. Data analysis was conducted using Microsoft Excel.

## RESULTS

In 2022, a total of 475,325 appointments were provided by public dental services in Porto Alegre. Patient profile showed a predominance of females (62%) and individuals aged 35 to 59 years (38%). Patients in early childhood (0 to 4 years) accounted for only 3% of the cases at primary care units, while older adults (60 years and older) accounted for 17%.

### Structure evaluation

In 2022, the profiles of 85 dentists (34%) from 61 primary care units (56%) were assessed through a questionnaire. It was found that 80% were women and 75% were under 40 years of age. Additionally, for 70% of the sample, time since graduation was less than fifteen years, and 78% had less than five years' experience in primary care. Over 60% of the dentists reported having a *lato sensu* specialization, while 18% reported a *stricto sensu* qualification. The most common specialization was public health.

### Process evaluation

The following actions and procedures are established

for primary care services<sup>11</sup>. These practices were categorized into three groups: type of care, active search for patients and dental actions and procedures, integrative practices and complementary medicine.

### Type of care

Overall, 71% of appointments were scheduled follow-ups, while 18% were first-time visits. Urgent dental care represented 7%, and 3% involved initial consultations and counseling. The remaining 1% consisted of other care types.

### Active Search for Patients

Telehealth within primary care constituted the most prevalent approach, accounting for 36% of the actions, followed by home visits, with 31%. Home care services constituted 24%, and 7% involved active searches.

### Dental Actions and Procedures

In Porto Alegre, primary care units conducted a total 422,107 registered dental procedures. Most of these procedures (n = 167,889; 40%) focused on Prevention and Health Promotion. Esthetic and functional rehabilitation corresponded to 21%, followed by Dental Urgency at 15%, Other Practices and Activities at 13%, and Oral Maxillofacial Surgery at 11%. Integrative practices accounted for less than 1%.

Table 1 presents the prevention and health promotion procedures performed in Porto Alegre in 2022. Oral hygiene counseling, prophylaxis, and scaling and root planing comprised more than 90% of all procedures. In contrast, topical fluoride application accounted for only 5%, while the use of pit-and-fissure sealants and cariostatic agents accounted for less than 1%.

Table 2 provides an overview of the esthetic and functional rehabilitation procedures performed in the city. Composite resin restorations on permanent teeth accounted for over 70% of the total procedures. In contrast, glass ionomer cement was the most commonly used restorative material for deciduous teeth. Silver amalgam restorations were infrequent, accounting for less than 1% of all procedures.

Table 3 presents the urgent dental procedures performed in Porto Alegre in 2022. Temporary cavity sealing (51%) and pulp access with intracanal medication (46%) comprised over 95% of the procedures in this category. The remaining 2% included other urgent interventions.

**Table 1. Dental actions and procedures for prevention and health promotion performed in primary care setting according to sex. Porto Alegre, Rio Grande do Sul, Brazil, 2022**

| Prevention and Health Promotion                                  | Male<br>n (%) |              | Female<br>n (%) |              | n (%)          |               |
|------------------------------------------------------------------|---------------|--------------|-----------------|--------------|----------------|---------------|
| <b>Total</b>                                                     | <b>61,968</b> | <b>(37%)</b> | <b>105,921</b>  | <b>(63%)</b> | <b>167,889</b> | <b>(100%)</b> |
| Oral hygiene guidance                                            | 31,030        | (37%)        | 51,821          | (63%)        | 82,851         | (49%)         |
| Scaling, root planing, and supragingival polishing (per sextant) | 14,914        | (36%)        | 26,548          | (64%)        | 41,462         | (25%)         |
| Prophylaxis / bacterial plaque removal                           | 5,915         | (37%)        | 10,108          | (63%)        | 16,023         | (10%)         |
| Subgingival scaling and root planing (per sextant)               | 3,707         | (38%)        | 6,149           | (62%)        | 9,856          | (6%)          |
| Topical fluoride application (individual per session)            | 3,293         | (38%)        | 5,331           | (62%)        | 8,624          | (5%)          |
| Coronal-root scaling (per sextant)                               | 1,093         | (39%)        | 1,698           | (61%)        | 2,791          | (2%)          |
| Prosthesis hygiene guidance                                      | 669           | (29%)        | 1,600           | (71%)        | 2,269          | (1%)          |
| Others                                                           | 1,347         | (34%)        | 2,666           | (66%)        | 4,013          | (2%)          |

**Table 2. Dental actions and procedures for esthetic and functional rehabilitation performed in primary care setting according to sex. Porto Alegre, Rio Grande do Sul, Brazil, 2022**

| Esthetic and Functional Rehabilitation                           | Male<br>n (%) |              | Female<br>n (%) |              | n (%)         |               |
|------------------------------------------------------------------|---------------|--------------|-----------------|--------------|---------------|---------------|
| <b>Total</b>                                                     | <b>33,801</b> | <b>(38%)</b> | <b>54,765</b>   | <b>(62%)</b> | <b>88,566</b> | <b>(100%)</b> |
| Restoration of posterior permanent tooth with composite resin    | 14,515        | (36%)        | 26,308          | (64%)        | 40,823        | (46%)         |
| Restoration of anterior permanent tooth with composite resin     | 9,044         | (40%)        | 13,486          | (60%)        | 22,530        | (25%)         |
| Pulp capping                                                     | 2,415         | (36%)        | 4,227           | (64%)        | 6,642         | (7%)          |
| Atraumatic restorative treatment (ART)                           | 2,269         | (41%)        | 3,313           | (59%)        | 5,582         | (6%)          |
| Occlusal adjustment                                              | 1,931         | (37%)        | 3,355           | (63%)        | 5,286         | (6%)          |
| Restoration of posterior primary tooth with glass ionomer cement | 1,923         | (48%)        | 2,078           | (52%)        | 4,001         | (5%)          |
| Restoration of posterior primary tooth with composite resin      | 891           | (48%)        | 954             | (52%)        | 1,845         | (2%)          |
| Others                                                           | 813           | (44%)        | 1,044           | (56%)        | 1,857         | (2%)          |

**Table 3. Urgent dental actions and procedures performed in primary care setting according to sex. Porto Alegre, Rio Grande do Sul, Brazil, 2022**

| Urgency                                                         | Male<br>n (%) |              | Female<br>n (%) |              | n (%)         |               |
|-----------------------------------------------------------------|---------------|--------------|-----------------|--------------|---------------|---------------|
| <b>Total</b>                                                    | <b>24,469</b> | <b>(38%)</b> | <b>39,287</b>   | <b>(62%)</b> | <b>63,756</b> | <b>(100%)</b> |
| Temporary sealing of dental cavity                              | 12,416        | (38%)        | 20,032          | (62%)        | 32,448        | (51%)         |
| Access to dental pulp and medicament (per tooth)                | 6,009         | (40%)        | 9,084           | (60%)        | 15,093        | (24%)         |
| Intracanal medicament with or without biomechanical preparation | 5,308         | (37%)        | 8,894           | (63%)        | 14,202        | (22%)         |
| Others                                                          | 736           | (37%)        | 1,277           | (63%)        | 2,013         | (3%)          |

Table 4 presents procedures classified as other practices and activities performed in the city. Approximately 60% of these actions involved non-dental procedures conducted by dentists. The most frequent procedures included the rapid SARS-CoV-2 detection test (39%), temperature and blood pressure measurement (30%), and

collecting samples for laboratory examination (16%), collectively accounting for over 80% of the total. Other practices and activities accounted for less than 1% (Table 4).

Table 5 presents the oral and maxillofacial surgical procedures performed in Porto Alegre in 2022. Approximately 95% involved permanent tooth

**Table 4. Other practices and activities performed in primary care setting according to sex. Porto Alegre, Rio Grande do Sul, Brazil, 2022**

| Other Practices and Activities                           | Male<br>n (%) |              | Female<br>n (%) |              | n (%)         |                 |
|----------------------------------------------------------|---------------|--------------|-----------------|--------------|---------------|-----------------|
| <b>Total</b>                                             | <b>20,815</b> | <b>(39%)</b> | <b>32,776</b>   | <b>(61%)</b> | <b>53,591</b> | <b>(100%)</b>   |
| <b>Rapid test for SARS-CoV-2 detection</b>               | 8,217         | (40%)        | 12,483          | (60%)        | <b>20,700</b> | <b>(39%)</b>    |
| <b>Measurement of temperature and blood pressure</b>     | 6,258         | (39%)        | 9,974           | (61%)        | <b>16,232</b> | <b>(30%)</b>    |
| <b>Collection of material for laboratory examination</b> | 3,254         | (38%)        | 5,316           | (62%)        | <b>8,570</b>  | <b>(16%)</b>    |
| <b>Rapid test for HIV/HBV/Syphilis/Hepatitis C</b>       | 991           | (39%)        | 1,544           | (61%)        | <b>2,535</b>  | <b>(5%)</b>     |
| <b>Anthropometric evaluation</b>                         | 781           | (37%)        | 1,309           | (63%)        | <b>2,090</b>  | <b>(4%)</b>     |
| <b>Clinical evaluation of smoker</b>                     | 625           | (34%)        | 1,216           | (66%)        | <b>1,841</b>  | <b>(3%)</b>     |
| <b>Intraoral X-ray</b>                                   | 513           | (41%)        | 726             | (59%)        | <b>1,239</b>  | <b>(2%)</b>     |
| <b>Others</b>                                            | 176           | (46%)        | 208             | (54%)        | <b>384</b>    | <b>(&lt;1%)</b> |

**Table 5. Oral-maxillofacial surgeries performed in primary care setting according to sex. Porto Alegre, Rio Grande do Sul, Brazil, 2022**

| Oral and Maxillofacial Surgery                  | Male<br>n (%) |              | Female<br>n (%) |              | n (%)         |               |
|-------------------------------------------------|---------------|--------------|-----------------|--------------|---------------|---------------|
| <b>Total</b>                                    | <b>19,126</b> | <b>(43%)</b> | <b>25,702</b>   | <b>(57%)</b> | <b>44,828</b> | <b>(100%)</b> |
| <b>Extraction of permanent tooth</b>            | 10,391        | (43%)        | 13,928          | (57%)        | <b>24,319</b> | <b>(54%)</b>  |
| <b>Removal of surgical stitches</b>             | 4,594         | (40%)        | 6,841           | (60%)        | <b>11,435</b> | <b>(26%)</b>  |
| <b>Extraction of primary tooth</b>              | 2,309         | (48%)        | 2,490           | (52%)        | <b>4,799</b>  | <b>(11%)</b>  |
| <b>Suture of injury in mouth and skin</b>       | 931           | (42%)        | 1,262           | (58%)        | <b>2,193</b>  | <b>(5%)</b>   |
| <b>Periapical curettage</b>                     | 251           | (45%)        | 304             | (55%)        | <b>555</b>    | <b>(1%)</b>   |
| <b>Odontosection / radiclectomy / tunneling</b> | 278           | (58%)        | 200             | (42%)        | <b>478</b>    | <b>(1%)</b>   |
| <b>Others</b>                                   | 372           | (35%)        | 677             | (65%)        | <b>1,049</b>  | <b>(2%)</b>   |

**Table 6. Integrative practices and complementary medicine performed in primary care setting according to sex. Porto Alegre, Rio Grande do Sul, Brazil, 2022**

| Integrative Practices/Complementary Medicine | Male<br>n (%) |              | Female<br>n (%) |              | n (%)        |               |
|----------------------------------------------|---------------|--------------|-----------------|--------------|--------------|---------------|
| <b>Total</b>                                 | <b>1,092</b>  | <b>(31%)</b> | <b>2,385</b>    | <b>(69%)</b> | <b>3,477</b> | <b>(100%)</b> |
| <b>Aromatherapy session</b>                  | 533           | (34%)        | 1,037           | (66%)        | <b>1,570</b> | <b>(45%)</b>  |
| <b>Chromotherapy session</b>                 | 422           | (36%)        | 755             | (64%)        | <b>1,177</b> | <b>(34%)</b>  |
| <b>Auriculotherapy session</b>               | 20            | (7%)         | 272             | (93%)        | <b>292</b>   | <b>(8%)</b>   |
| <b>Phytotherapeutic treatment</b>            | 78            | (31%)        | 175             | (69%)        | <b>253</b>   | <b>(7%)</b>   |
| <b>Naturopathic treatment</b>                | 25            | (32%)        | 52              | (68%)        | <b>77</b>    | <b>(2%)</b>   |
| <b>Hands-on healing session</b>              | 7             | (10%)        | 63              | (90%)        | <b>70</b>    | <b>(2%)</b>   |
| <b>Other</b>                                 | 7             | (18%)        | 31              | (82%)        | <b>38</b>    | <b>(1%)</b>   |

extractions (54%), suture removal (31%), and deciduous tooth extractions (11%). The remaining 2% comprised other surgical interventions.

Integrative Practices and Complementary Medicine Table 6 presents the integrative practices and complementary medicine procedures performed by dentists in Porto Alegre in 2022. Aromatherapy and chromotherapy sessions comprised over 70% of

these practices, while other integrative approaches accounted for only 1%.

## DISCUSSION

The integration of oral health into Brazil's public healthcare system is a relatively recent development, with policies designed to align actions and services with the actual health needs of the population<sup>12</sup>.

To enhance health outcomes, various assessment tools have been employed to evaluate structural components and work processes, aiming to improve indicators among system users<sup>13</sup>.

This study identified that most oral healthcare services are provided to women aged 20 to 59 years, while early childhood patients are underrepresented. Similarly, Cassal et al.<sup>1</sup> found that 77% of users in Porto Alegre were women aged 20 to 50 years. Other studies suggest that women frequently assume the role of seeking healthcare for their families<sup>14, 15</sup>. Despite recommendations for the first dental visit to occur within the first year of life<sup>16</sup>, dental appointments for infants remain uncommon, in agreement with previous studies<sup>17, 18</sup>. A local cohort study associated early dental visits with higher maternal education and socioeconomic status but found no association with the number of professionals or team structure<sup>19</sup>.

The profile of primary care dentists in Porto Alegre revealed a predominance of women with specialization in collective health. These findings are supported by D'Avila et al.<sup>20</sup>, who reported similar characteristics using the PCATool-Brazil, and by national data from the PMAQ, indicating that 57% of surveyed dentists had postgraduate training in collective health<sup>8</sup>. This educational background has a positive impact on team performance and highlights the importance of ongoing training in primary care<sup>7</sup>.

In terms of service organization, most visits were scheduled return appointments, suggesting structured care. A flexible appointment system, as discussed by Mendes et al.<sup>7</sup>, is associated with better team performance. Nonetheless, urgent visits—although comprising only 5%—remain essential for low-income populations, who often seek care only when in pain and tend to access services without prior scheduling<sup>1</sup>. Among outreach strategies, telehealth was the most frequently used. Widely adopted during the COVID-19 pandemic, this modality was encouraged by the Rio Grande do Sul state government<sup>21</sup>. Internationally, tele-screening proved effective for emergency triage<sup>22</sup> and postoperative follow-up<sup>23</sup>. In Brazil, although Resolution 226/2020 initially restricted remote dental care<sup>24</sup>, exceptions were made for tele-monitoring and tele-counseling, especially in prenatal care<sup>25</sup>. Since then, “tele-appointments” have been incorporated into primary care routines, supporting active patient engagement<sup>25, 26</sup>.

An analysis of dental procedures and interventions

revealed strong emphasis on prevention and health promotion. These procedures were frequent, and include for example oral hygiene guidance and supragingival scaling, which are effective in periodontal disease prevention and meet most primary care demands<sup>27</sup>. However, the use of preventive agents such as sealants, cariostatic agents and fluoride remains limited, despite strong recommendations from Pediatric Dentistry associations<sup>28, 29</sup>. High demand often leads professionals to prioritize curative and rehabilitative treatments<sup>28, 29</sup>.

Composite resin was the most used material for restorations in permanent teeth, especially in posterior regions. This preference is probably due to the material's esthetic appeal, conservative technique and reparability<sup>30, 31</sup>. In contrast, silver amalgam was rarely used, reflecting concerns about its appearance and mercury content<sup>31, 32</sup>.

Atraumatic Restorative Treatment (ART) is a cost-effective, minimally invasive strategy suitable for both public and private sectors. Its benefits include simplified technique, tissue preservation and behavioral management<sup>33</sup>. Given its low utilization, we recommend its inclusion in continuing education initiatives to promote broader adoption in primary care<sup>34</sup>.

Urgent procedures were mostly driven by pain of pulpal origin from deep cavities, aligning with previous reports<sup>1</sup>. In these cases, pulpal access and intracanal medication are crucial to relieve symptoms<sup>32</sup>. Nevertheless, more than half of surgical procedures involved extractions. Delays in endodontic treatment – up to four years in secondary care in Porto Alegre – contribute to this outcome<sup>35</sup>, along with limited access to diagnostic tools and materials in primary care<sup>36</sup>.

Integrative and complementary practices, regulated since 2006<sup>37</sup>, have gained space in dental care to support anxiety and stress reduction. Techniques such as aromatherapy, chromotherapy and auriculotherapy were reported in this study. While some evidence supports their benefits in conditions such as temporomandibular disorders and bruxism<sup>38, 39</sup>, there is still a lack of high-quality studies to justify broader public investment.

Lastly, procedures not traditionally associated with dentistry, such as SARS-CoV-2 testing, vital sign measurement, and sample collection, were also reported. These practices emerged during the COVID-19 pandemic, when elective dental care was restricted,

and dentists were repurposed for broader health support roles<sup>40</sup>. Their continued execution reflects the ongoing impact of the pandemic on dental practice.

While the study highlights an organized dental care structure within the public system, the findings show that the services do not fully align with Primary Health Care principles, particularly in terms of relevance, appropriateness and life-course suitability. The underuse of services by young children, the focus on curative rather than preventive care, and the limited evidence-based strategies, such as atraumatic restorative treatment (ART) and fluoride, reveal gaps between service provision and population needs. To improve oral health in the universal healthcare system (*Sistema Único de Saúde*, SUS), actions should target life-course needs and age-specific demands, especially in early childhood.

This study is limited by the reliability of secondary data, which may lead to over- or underestimation of certain indicators. Additionally, the cross-sectional design

precludes causal inferences. However, its strength lies in the regional scope, offering relevant insights into the organization of primary oral health care in a major city in southern Brazil. The findings may inform improvements in service delivery and planning aligned with the principles of primary health care.

## CONCLUSIONS

The present study highlighted key aspects of the structure and processes of oral healthcare within primary care settings in Porto Alegre, emphasizing the need for ongoing educational initiatives for healthcare providers. These initiatives should focus on integrating proven, yet underutilized, procedures such as atraumatic restorative treatment and pit-and-fissure sealants. Additionally, there is an urgent need to address early childhood dental care and the use of technologies in primary care as essential components of comprehensive oral health management.

## CONFLICT OF INTERESTS

The authors declare no potential conflicts of interest with regards to the study, authorship, and/or publication of this article.

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# Effect of mechanized instrumentation on distal wall thickness of the second mesiobuccal canal

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## ABSTRACT

Dentine wall thickness in the mesial roots of molars is critical to prevent root stripping. Understanding the effect of rotatory instruments on the second mesiobuccal canal is crucial to prevent weakening and periodontal communication. **Aim:** To evaluate the effect of different conservative tapered mechanized instrumentation systems on the distal wall thickness of the second mesiobuccal canal of maxillary first molars in 3D dental replicas. **Materials and Method:** Fifty radiopaque 3D printed replicas were made of a maxillary first molar with Vertucci classification IV and Schneider curvature ( $<10^\circ$ ), 23 mm in length. The specimens were divided into 5 groups of 10. In all cases, the second mesiobuccal canal was instrumented to the working length. Group 1 was instrumented with Race Evo (FKG) up to file 30/04, Group 2 with AF F One (Fanta-Dental) up to file 35/04, Group 3 with Slim Shaper (Zarc4Endo) up to file 30/04, and Group 4 with Trunatomy, while the control group was not instrumented. Distal wall thicknesses were measured in the cervical, middle and apical thirds by CBCT before and after instrumentation. Descriptive statistics were calculated, after which analysis of variance and Tukey's test for multiple comparisons were performed ( $p < 0.05$ ). **Results:** Analysis of variance showed a significant effect of instrumented group and thirds ( $p < 0.001$ ). The multiple comparison test showed that the control and Race Evo groups differed significantly from the other three groups. Within each group, the differences between thirds were statistically significant. **Conclusion:** Trunatomy, Slim Shaper and Fanta AF One systems are suitable for shaping second mesiobuccal canals while maintaining critical distal wall thickness.

**Keywords:** endodontics - root canal preparation - molar

## Efecto de la instrumentación mecanizada sobre el espesor de la pared distal del segundo conducto mesiovestibular

## RESUMEN

El espesor de la pared distal en las raíces mesiales de molares es crítico para evitar perforaciones radiculares. Conocer el efecto de los instrumentos rotatorios sobre el segundo conducto mesiovestibular resulta crucial para evitar su debilitamiento y comunicación periodontal.

**Objetivo:** Evaluar el efecto de diferentes sistemas de instrumentación mecanizada de conicidades conservadoras en el espesor de la pared distal de segundo conducto mesiovestibular de primeros molares superiores en réplicas dentarias 3D. **Materiales y Método:** Se confeccionaron 50 réplicas 3D radiopacas de un primer molar superior con una clasificación IV de Vertucci y con grado de curvatura de Schneider leve ( $<10^\circ$ ), de 23 mm de longitud y se dividieron 5 grupos de 10 muestras. En todos los casos se instrumentó el conducto segundo conducto mesiovestibular MB2 hasta la longitud de trabajo. El Grupo 1 fue instrumentado con Race Evo (FKG) hasta lima 30/04, Grupo 2 instrumentado con AF F One (Fanta- Dental) hasta lima 35/04, Grupo 3 con Slim Shaper (Zarc4Endo) hasta lima 30/04, Grupo 4 con Trunatomy (Densply Sirona) hasta lima 36/03 y el grupo testigo sin instrumentar. Se midieron los espesores dentinarios de la pared distal antes y posterior a la instrumentación de los sistemas mediante tomografía computarizada de haz cónico. Luego del cálculo de los estadísticos descriptivos se realizó el análisis de varianza y las comparaciones múltiples se realizaron por medio de la prueba de Tukey con una  $p$  de 0.05. **Resultados:** El análisis de variancia muestra el efecto significativo de los factores grupo instrumentado y tercio ( $p < 0.001$ ). La prueba de comparación múltiple muestra diferencias estadísticamente significativas entre los grupos control y Race Evo y los otros tres. Dentro de cada grupo las diferencias entre los tercios son estadísticamente significativas. **Conclusión:** Los sistemas Trunatomy, Slim Shaper y Fanta AF One son adecuados para la conformación del CMB2 manteniendo el espesor crítico de la pared distal.

**Palabras clave:** endodoncia - preparación del conducto radicular - molar

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## INTRODUCTION

Detection and adequate treatment of the second mesiobuccal (MB2) canal in the mesial root of first upper molars are essential to ensure the success of endodontic treatment<sup>1</sup>.

A cross section of a molar mesial root shows that its thickest part (safety zone) is away from the direction of its curvature, while the concave interior wall, called Risk/Danger Zone, is located on the distal dentin wall facing the furcation zone in upper and lower molars<sup>2</sup>.

“Stripping” is the thinning and perforation of the distal wall of mesial roots as a result of mechanical shaping using excessively tapered instruments. These unwanted perforations are followed by bacterial contamination, periradicular tissue lesion, periodontal inflammation, bone resorption, periodontal ligament destruction, gingival epithelium proliferation and periodontal pocket development<sup>3</sup>.

Prakash et al.<sup>4</sup> reported that the diameter of the MB2 canal is smaller and usually narrower than in the first mesiobuccal canal (MB1). In MB2 canals, the thickness of the distal dentin wall is one of the most important factors to consider before instrumentation. The longer the root canal, the more likely it is to undergo root perforation by stripping, because at the distal wall, the dentin is thinner, and the concavity is more pronounced than in shorter molars<sup>5</sup>.

Mechanical shaping of root canals can lead to various degrees of dentin removal, depending on the instrumentation techniques and differently tapered mechanized systems used. This can affect the biomechanical response of teeth and weaken their capacity to bear occlusal loads in the long term, leading to tooth loss as a result of excessive wear of distal dentin in mesial roots.

The thickness of the risk zone before and after instrumentation has been measured on extracted teeth using different methods, including tomographic studies<sup>6</sup>, micro-CT scan<sup>7</sup> and stereomicroscopy<sup>8</sup>. However, it is often difficult to obtain extracted teeth for *ex-vivo* studies. An alternative solution is to use 3D dental replicas instead.

Extracted natural teeth have certain disadvantages in research compared to 3D replicas, e.g., the time and difficulty involved in finding enough natural teeth to make up a representative sample<sup>9-11</sup>. Another factor to consider is that patients must provide informed consent for the use of natural extracted

teeth because they are biological tissue<sup>12</sup>. Moreover, there is a risk of cross-infections if teeth are contaminated. Natural extracted teeth are difficult to sterilize, and common disinfection procedures such as sodium hypochlorite and hydrogen peroxide may destroy them<sup>13-14</sup>. Furthermore, the difficulty in standardizing anatomical evaluations because of the complex anatomy of the root canal system makes it impossible to obtain completely uniform models for scientific studies<sup>15-16</sup>.

3D replica printing is a rapidly developing technology that has gained broad acceptance in dentistry and can be used to resolve these difficulties<sup>17</sup>. In a recent study, Ramírez-Muñoz A et al.<sup>18</sup> demonstrated the importance of 3D printed models for endodontic research thanks to the precision with which they faithfully replicate specific anatomy of natural teeth, providing a standardized alternative providing specimens which are much easier to obtain than natural teeth.

The aim of this study was to evaluate and compare the effect of endodontic instrumentation with different mechanized systems in 3D replicas of first upper molars, focusing on the preservation of the MB2 canal's distal wall thickness.

## MATERIALS AND METHOD

A preliminary pilot study was conducted to determine the arithmetic mean of distal wall thicknesses in the cervical, middle and apical thirds of the second mesiobuccal canal (MB2).

We evaluated tomographic images from a database of 137 MB2 canals from mesial roots of 225 first upper molars of patients (mean age 29 ±5.4 years), which had been taken at a dental radiology center specializing in CBCT. Measurements were taken with the calibration software of a Carestream CS 9600 3D scanner with image acquisition parameters established at 82 kV and 8.0 mA, exposure time 19.96 s. Minimum voxel size was 180 µm. The thickness of the distal wall of the second mesiobuccal canal was measured in the cervical, middle and apical thirds. Before this step, the length of each root was measured from the furcation to the root apex in order to divide it into three parts. The arithmetic means were 1 mm for the cervical third, 0.76 mm for the middle third and 0.56 mm for the apical third.

Based on the dentin thicknesses found, a first molar that was representative of the mean value was selected from the DICOM image bank of the patients

evaluated: an upper first molar with MB2 canal, Vertucci classification IV and Schneider curvature  $<10^\circ$ . This molar was replicated physically in 3D anatomical models.

Sample size was calculated with the Software IBM SPSS Statistics Version 24 (IBM Corp, Endicott, USA), establishing an alpha error 0.05 and 95% power. The result indicated a minimum of 10 specimens per study group.

Thus, fifty (50) radiopaque replicas (LikeReal 3D [Porto Alegre, Rio Grande do Sul, Brazil]) were made of the selected molar. These replicas had pre-made openings, 23 mm root length to the apical foramen, and the same dentin thicknesses in each third as the selected molar (Fig. 1).

The 3D replicas were divided into four (4) experimental groups in which the MB2 canal would be prepared with different mechanized instrumentation systems for evaluation of residual dentin thickness (Fig. 1), and one (1) control group without instrumentation. The replicas were placed on an acrylic plaque and scanned in the same scanner with image acquisition parameters 90 kV, 4.0 mA, voxel size  $0.75\ \mu\text{m}$  and exposure time 20 seconds. The DICOM images were used to measure the furcal dentin thicknesses in the MB2 canals in the cervical, middle and apical thirds (Figs. 2 - 5).



Fig. 1: Vestibular view of 3D replica of upper first molar.



Fig. 2: 3D tooth replicas on a plaque, ready to scan.

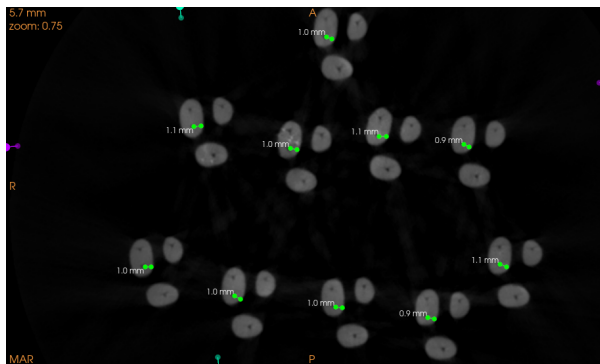


Fig. 3: Measurement of the distal wall in the cervical third of the control group.

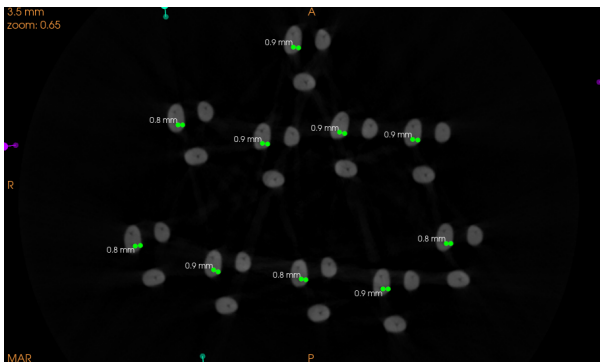


Fig. 4: Measurement of the distal wall in the middle third of the control group.

After recording the measurements, and before shaping, patency was confirmed by inserting a file #10 (Dentsply Maillefer - Ballaigues, Switzerland) up to the 23 mm working length.

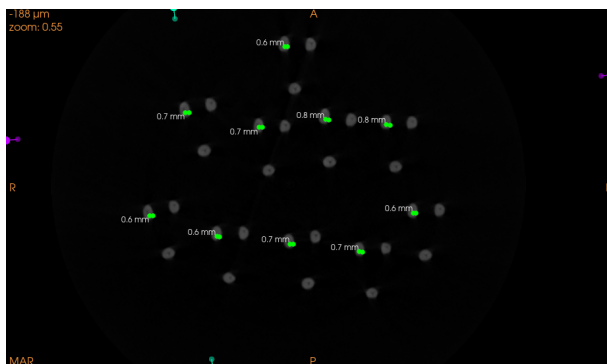


Fig. 5: Measurement of the distal wall in the apical third of the control group.

Each rotary instrument was used to instrument a maximum of four MB2 canals, following the recommendations of the manufacturers of each system.

The rotary file systems were driven by an endodontic motor (Eighteenth - Changzhou, China). In-and-out movements with an amplitude of 3 to 4 mm were made until the working length was reached. Brushing movements were not used at any time. After use, the instruments were cleaned with sterilized gauze and alcohol.

For all experimental groups, root canals underwent an irrigation protocol with 2.5 % sodium hypochlorite using a 21 mm NaviTip 29G needle (Ultradent- Utah, USA) placed at 3 mm from the working length. Four (4) ml of the solution were used between each instrument change and at the end of instrumentation, plus a final irrigation protocol with 10 ml.

All specimens were instrumented by a single operator (P.E.), who is a specialist in endodontics and experienced in continuous rotary instrumentation, using an endodontic Newton MEC XXI microscope (Newton Microcopia, Buenos Aires, Argentina) at 4 X magnification.

Experimental groups were the following:

**Group 1:** The Race-Evo (RE) system (FKG Dentaire, La Chaux-de-Fonds, Suiza) was used in continuous rotation at 1000 rpm, torque 1.5 N/cm, following the manufacturer's instructions. When the working length was reached, the file was removed from the root canal, and the protocol continued using the following sequence of files: RE1 (15/.04), RE2 (25/.04) and RE3 (30/.04).

**Group 2:** The AF F ONE (AF) system (FANTA-DENTAL, Shanghai, China) was used at 500 rpm, torque 2.5 N/cm. When the working length

was reached, the protocol continued using the file sequence recommended by the manufacturer: AF 20/.04, 25/.04 and 35/.04.

**Group 3:** The Slim Shaper (SS) system (Zarc4endo, Gijón, Asturias, Spain) was used with continuous rotary speed 500 rpm, torque 3 N/cm. When working length was reached, the protocol continued using the file sequence recommended by the manufacturer: ZS1 (15/.02–06), ZS2 (20/.04) and ZS3 (25/.04). Finally, the apical third was widened with the instrument AS 30 (30/.03) from Apical Shaper (AS) (Zarc4endo, Gijón, Asturias, Spain) at 500 rpm, torque 3 N/cm.

**Group 4:** The TruNatomy (TA) system (Dentsply Sirona, Ballaigues, Switzerland) was used at continuous rotary speed 500 rpm, torque 1.5 N/cm. When working length was reached, the protocol continued using the file sequence specified in the manufacturer's instructions: TA Glyder (17/.02), Prime (26/.04) and Medium (36/.03).

After all the replicas had been instrumented, the same CBCT scanner was used to scan them with the same image acquisition parameters, to evaluate distal dentin wall thickness remaining after instrumentation.

We evaluated the differences among all five groups (four study groups and control group), and the differences in thickness among root thirds.

After calculating the descriptive statistics (arithmetic mean, standard deviation, and maximum and minimum values for each group of ten data each), two-way ANOVA was performed (group according to instrument and third considered, the latter with repeated measures) after establishing that the homogeneity of variance hypothesis could be accepted for the dependent variable between experimental groups. Multiple comparisons were made through Tukey's test, with significance level set at a probability value (alpha) lower than 0.05 in all cases.

## RESULTS

Table 1 shows the descriptive statistics (arithmetic mean, standard deviation, and maximum and minimum values for each group).

Analysis of variance shows the significant effect of the factors "instrumented group" and "third" ( $p < 0.001$ ). Interaction is significant, but at a lower level (0.041) (Table 2).

The global multiple comparison test (considering total thirds within each group) shows that the control

**Table 1. descriptive statistics of thicknesses in the experimental groups of molar replicas. Values expressed in mm.**

| Group     | Third    | n  | Mean | S.D. | Minimum | Maximum |
|-----------|----------|----|------|------|---------|---------|
| Control   | CERVICAL | 10 | 1.01 | 0.07 | 0.9     | 1.1     |
|           | MIDDLE   | 10 | 0.86 | 0.05 | 0.8     | 0.9     |
|           | APICAL   | 10 | 0.68 | 0.08 | 0.6     | 0.8     |
| Fanta     | CERVICAL | 10 | 0.71 | 0.07 | 0.6     | 0.8     |
|           | MIDDLE   | 10 | 0.54 | 0.08 | 0.4     | 0.6     |
|           | APICAL   | 10 | 0.37 | 0.07 | 0.3     | 0.5     |
| Race Evo  | CERVICAL | 10 | 0.63 | 0.05 | 0.6     | 0.7     |
|           | MIDDLE   | 10 | 0.43 | 0.05 | 0.4     | 0.5     |
|           | APICAL   | 10 | 0.31 | 0.06 | 0.2     | 0.4     |
| Slim      | CERVICAL | 10 | 0.80 | 0.08 | 0.7     | 0.9     |
|           | MIDDLE   | 10 | 0.60 | 0.08 | 0.5     | 0.7     |
|           | APICAL   | 10 | 0.40 | 0.08 | 0.3     | 0.5     |
| Trunatomy | CERVICAL | 10 | 0.75 | 0.05 | 0.7     | 0.8     |
|           | MIDDLE   | 10 | 0.56 | 0.07 | 0.5     | 0.7     |
|           | APICAL   | 10 | 0.37 | 0.07 | 0.3     | 0.5     |

**Table 2. Summary of analysis of variance. Interaction is significant, but at a lower level (0.041)**

| Factor        | gl | Sum of squares | Mean squares | F       | P      |
|---------------|----|----------------|--------------|---------|--------|
| Group         | 4  | 2.647          | 0.662        | 64.832  | <0.001 |
| Third         | 2  | 3.134          | 1.565        | 763.635 | <0.002 |
| Group x third | 8  | 0.035          | 0.004        | 2.128   | 0.041  |

group and Race Evo differ significantly from the other three (Table 3).

Within each group, the differences between thirds are statistically significant ( $p < 0.05$ ), with values decreasing from cervical to apical.

## DISCUSSION

Remaining dentin thickness after endodontic procedures may be the most important factor influencing the future resistance of an endodontically treated tooth, so in the MB2 canal, it is important to consider it during instrumentation in order to avoid perforating the danger zone<sup>19</sup>.

This study evaluated the influence of four sixth-generation mechanized instrument systems with conservative taper on the wear of distal wall dentin, by means of standardized 3D replicas of upper molars. Significant differences were found in the remaining thickness of dentin/resin before and after instrumentation with the reduced taper systems.

**Table 3. Global Tukey's test (differences between groups in the same subset are not statistically significant). Values are the global arithmetic mean.**

| Group     | Subset |      |      |
|-----------|--------|------|------|
| Race Evo  | 0.46   |      |      |
| Fanta     |        | 0.54 |      |
| Trunatomy |        | 0.56 |      |
| Slim      |        | 0.60 |      |
| Control   |        |      | 0.85 |

Dentin wear as a result of root canal instrumentation is not a simple phenomenon, but the outcome of complex multifactorial interaction which includes:

1. Type of metallurgy: Heat treatments determine an instrument's main mechanical properties. They control the transition of phases between the crystalline structures of austenite (more rigid) and martensite (more ductile and flexible), which in turn determines the instrument's flexibility, resistance to cyclic fatigue and cutting efficiency<sup>20</sup>.
2. Instrument geometry: A file's macro- and micro-geometric design is a critical determinant of its clinical behavior. The instrument's cross section, taper, pitch, helicoidal angle and core diameter all directly influence stress distribution, cutting capacity, debris extrusion and instrument safety<sup>21-22</sup>.
3. Kinematics and Operational Parameters: The way in which the instrument is used, including rotational speed (rpm), torque and type of movement (continuous rotation or reciprocating), has a profound impact on its performance and safety. These parameters modulate the generation of stress both in the instrument and in the walls of the root canal, affecting cyclic fatigue, formation of dentin microcracks, and wear on the dentin wall<sup>23-24</sup>.

Although a definitive minimum dentin thickness of the remaining radicular wall has not yet been established scientifically, 0.3 mm is considered critical for bearing the masticatory impact<sup>25</sup>. In the current study, the arithmetic means of remaining thicknesses of the distal wall post-instrumentation were 0.72 mm in the cervical third, 0.53 mm in the middle third, and 0.36 mm in the apical third with instruments with maximum tapers of 0.04, suggesting that this taper may be safe for maintaining a dentin thickness able to withstand masticatory impact.

The rotary systems evaluated had similar tapering and diameters. Although the Race-Evo system generating significantly greater dentin/resin wear than Trunatomy, AF F One and Slim Shaper, this does not necessarily mean that the differences are clinically significant. The differences in the statistical results may be due to characteristics of the model used for this experiment and the rotary speed established by the manufacturer.

Vertucci type IV classification was selected for the 3D replicas because it is the most common arrangement of root canal system and with independent root canals. This enables a separate analysis of instrumentation effectiveness, and increases the size of the study sample<sup>26</sup>. Moreover, these canals are usually narrow, and the dentin on the distal wall of the mesial root is thin, which makes it interesting for evaluating dentin reduction and unprepared zones<sup>27</sup>.

The mechanical properties of 3D resin replicas differ from those of natural dentin. The Young module and maximum tensile strength of the resin used for these replicas are lower than those of human dentin, in which Young's module is approximately 18 GPa<sup>17</sup>. This resin, with a wavelength range of 365–405 nm, probably has a modulus of elasticity closer to 2–3 GPa, i.e., which makes it considerably softer and more flexible than natural dentin.

The rotary speed of the Race Evo system suggested by the manufacturer and used in this study (1,000 rpm) was double that used in the other systems. It may have generated heat by friction which could have softened the resinous material of the replicas, causing greater wear in the wall studied. Although 3D replicas are reliable along most of the length of the root canal, future studies could investigate whether alternative resins with properties closer to the mechanical profile of dentin could improve precision in studies on shaping, especially in anatomically complex areas, and determine whether heat caused by rotation softens the resin used. This information could be used to improve dental replica materials. This is one of the limitations of this study. The cross section of machined instruments is a critical factor influencing their cutting efficiency. Designs with smaller central mass, such as triangular sections, tend to be more flexible and produce very good cuts<sup>28</sup>. More innovative designs, such as off-centered parallelogram or asymmetrical cross sections, alter the dynamics of rotation to reduce continuous contact with the root canal walls<sup>22</sup>. This

reduction in contact reduces torsional stress on the instrument and, importantly, minimizes the dentinal wear caused by friction<sup>28</sup>.

The Fanta AF One (AF-R Wire, Blue) system uses “AF-R” wire technology with the aim of improving both the flexibility and the resistance of the instrument. The Race Evo system uses a surface electropolishing treatment that provides resistance to torsion<sup>29</sup>. Slim Shaper is based on a concept of “3 alloys”: a *Gold* alloy for maximum cutting capacity and torsional resistance, a *Pink* alloy providing a balance between cutting and flexibility, and a *Blue* alloy for extraordinary flexibility and high cyclic fatigue resistance<sup>30</sup>. Trunatomy instruments are made of small-diameter Gold alloy NiTi wire, with maximum diameter at D16 (MFD) of only 0.8 mm, compared to the 1.2 mm of many generic systems<sup>31</sup>. Instrument cross section is an important factor that influences cutting efficiency. Designs with smaller central mass, such as triangular sections, tend to be more flexible and cut well<sup>27</sup>. More innovative designs, such as off-centered parallelogram or asymmetrical cross sections, alter the dynamics of rotation to reduce continuous contact with the root canal walls<sup>22</sup>. This reduction in contact reduces torsional stress on the instrument and importantly, minimizes dentin wear due to friction<sup>27</sup>. In addition to influencing cutting efficiency, instrument cross section influences debris elimination capacity, flexibility and stress distribution<sup>21</sup>. Trunatomy has an off-centered parallelogram cross section that generates asymmetric rotation within the root canal, thereby reducing the contact area between the file and the root canal walls at any given time. Fanta AF One has an S-shaped cross section that provides two active cutting points which increase its cutting efficiency<sup>32</sup>. In contrast, Slim Shaper and Race Evo both have flat triangular cross sections.

Some studies highlight the ability of Trunatomy to preserve root canal anatomy, with minimal transportation<sup>33-34</sup>. Khanderparkar et al.<sup>35</sup> compared the machined systems XP-endo Shaper, TruNatomy and HyFlex CM, reporting that Trunatomy eliminated significantly less dentin at all levels measured, even though all three systems have the same taper (0.04). This may have been because Trunatomy has a regressive type taper. In contrast, Vadera et al.<sup>36</sup>, in a study using CBCT, found that Trunatomy eliminated significantly more dentin at 3 mm and 7 mm from the apex, compared to the 2Shape y One Curve systems. This discrepancy

might be attributable to differences in methodological evaluation between the two studies (one was evaluated by CBCT and the other by micro-CT), or to the specific characteristics of the systems compared.

In some studies that made direct comparisons, Race Evo performed better than Fanta AF F-One regarding dentin preservation in the central and apical regions of the root canal<sup>20,32</sup>. However, in the current study, Race Evo caused the greatest dentin/resin wear.

The Slim Shaper system caused the least wear in all three thirds. This is probably due to its flat triangular cross section, MDF 0.80, and regressive taper. Although the Race Evo system also has a flat triangular cross section and MDF lower than 1, its taper is constant, which may also have had an influence on the greater wear recorded in the current study.

While alloy properties and cross section influence flexibility and cutting efficiency, tapering is key to the instrument's potential to eliminate debris. Thus, the influence of the type and percentage of tapering may also be important to preserving dentin on the distal wall of the MB2 canal. This firmly supports the use of 0.04 tapers or regressive designs, which would minimize the risk of over-instrumentation and stripping the distal dentin wall of MB2 root canals.

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The results of the current study on shaping MB2 canals suggest that it may be appropriate to use rotary systems with conservative tapering, along with improved irrigation protocols, rather than using machined instruments with large taper, which could weaken the root's dentin support.

## CONCLUSIONS

Within the limitations of this study, it can be concluded that the Trunatomy, Slim Shaper and Fanta AF One systems are adequate for shaping the MB2 canal, preserving the critical thickness of the distal wall following endodontic instrumentation.

## CONFLICT OF INTERESTS

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

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