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Original Article

Shaping ability of three heat-treated NiTi systems in Vertucci's type III root canals of mandibular incisors: An *ex vivo* studyRenata Maira de Souza Leal^{a,*}, Felipe Andretta Copelli^a, Jáder Camilo Pinto^b,
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ABSTRACT

Introduction: The anatomical configuration classified as Vertucci's type III is described as the second most prevalent in mandibular incisors.**Methods:** Thirty-six Vertucci's type III mandibular incisors were evaluated by micro-computed tomography (micro-CT) and divided into 3 groups (n = 12) according to the root canal preparation protocol (HyFlex CM [HCM], HyFlex EDM [HEDM], and Sequence Rotary File [SRF]). The teeth were scanned before and after performing 0.25 mm and 0.40 mm apical diameter preparations. The canal volume, dentin thickness, percentage of accumulated debris and untouched canal areas, transportation, and centering ability were measured. The data were statistically analyzed by ANOVA, Tukey, Kruskal-Wallis, and Dunn tests (P < 0.05).**Results:** The volume increase was more evident in the apical third. After 0.40 mm preparation, the SRF system provided a higher reduction (P < 0.05) in dentin thickness on the buccal surface 1 mm from the apex. There was higher canal transportation in the bucco-lingual direction. The 0.40 mm apical preparation reduced the percentage of untouched canal areas. The apical third had the highest percentage of untouched canal areas. The cervical third had the lowest volume of accumulated debris.**Conclusions:** Increasing the apical preparation to a diameter of 0.40 mm with the HCM, HEDM, and SRF systems in Vertucci's type III root canals of mandibular incisors proved to be safe and effective, reducing untouched canal areas.**Clinical relevance:** Root flattening can be intense to the point of generating a root canal bifurcation. Despite the decrease in the root canal diameter, a greater enlargement of the apical region is necessary and safe.

1. Introduction

It is essential to maintain the original characteristics of the canal and dentin as much as possible while preparing the root canal (De-Deus et al., 2019; Schilder, 1974). However, a greater enlargement of the apical diameter has been shown to have a significantly positive impact on canal cleaning, resulting in more effective removal of debris (De-Deus et al., 2015) and better disinfection of the root canal. Moreover, this enlargement allows a deeper penetration of the irrigation needle and the usage of larger volumes of the irrigating chemical solution (Brunson et al., 2010), independently of the irrigating solution employed

(Rodrigues et al., 2017).

The presence of anatomical complexities can increase the challenges associated with root canal preparation, often leading to areas within the root canal system that remain untouched (Siqueira Junior et al., 2018). Several studies have shown the unsuccessful efficiency of various systems in preparing oval-shaped canals of mandibular incisors (Azim et al., 2017; Pérez et al., 2018; Velozo et al., 2020; Versiani et al., 2017; Zuolo et al., 2018). These studies reported that up to 58.8 % of the canal space remains untouched (Azim et al., 2017).

Vertucci's type III (Vertucci, 1984) is the second most prevalent anatomical configuration in mandibular incisors, with prevalence

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ranging from 0.6 % to 33.0 % (Leoni et al., 2014; Martins et al., 2018; Milanezi de Almeida et al., 2013; Shemesh et al., 2018; Thomas et al., 2020). The mean apical diameter in this configuration is approximately 0.41 mm in the bucco-lingual direction and 0.22 mm in the mesio-distal direction (Milanezi de Almeida et al., 2013). Despite this prevalence, there is a current lack of research focused on the preparation of root canals with this anatomical condition.

Therefore, our research aimed to assess the effectiveness of three rotary systems with different heat treatments in shaping mandibular incisor root canals classified as Vertucci's type III up to different levels of apical enlargement.

The null hypotheses of the study are: (i) Apical enlargement up to the instrument tip diameter of 0.40 mm will not compromise the dentin structure, regardless of the system used. (ii) There is no difference in the shaping ability among the HCM, HEDM, and SRF systems.

2. Methods

2.1. Sample selection

The sample size calculation was performed using the G*Power v3.1 software for Mac OS (Heinrich Heine, Universität Düsseldorf, Germany) and the ANOVA test, considering the alpha type error at 0.05, beta power of 0.80, and expected effect of 1.1, based on the studies of [Vertuciani et al. \(2018\)](#) and [Zuolo et al. \(2018\)](#). We obtained a total sample of 36 teeth ($n = 12$).

The experimental study was approved by the local Ethics Committee (protocol number 1.051.377). Extracted human mandibular incisors from a local human tooth bank were used. The inclusion criteria were teeth with single root and complete rhizogenesis, without previous endodontic treatment, and with intense root flattening in the mesio-distal direction. The teeth were radiographed by digital radiography (Kodak RVG 500, Eastman Kodak, USA) in the mesio-distal direction, and those with the suggestion of root canal bifurcation were selected. Then, the samples were scanned using the Skyscan 1174 micro-CT (Bruker-microCT, Kontich, Belgium) with parameters of 50 kV, 800 μ A, rotation angle of 0.7, and voxel size of 16.8 μ m. The obtained images were reconstructed using the NRecon v.1.6.4.8 software (Bruker-microCT) using the same parameters for all teeth. The reconstruction of three-dimensional models and evaluation of the volume and diameter of the root canals were performed using the CTAn v.1.12 and CTVol v.2.2.1 softwares (Bruker-microCT). The volume (mm^3) of the canal was measured in three segments: 1–4 mm (apical third), 4–7 mm (middle third), and 7–10 mm (cervical third) from the apical foramen. The minor and major diameters of the buccal and lingual canals were measured in five segments: 1 mm from the apical foramen, in the cemento-enamel junction (CEJ), at the beginning of the canal bifurcation, in the middle sections of the buccal and lingual canals, and at the joining of the canals. Therefore, 36 mandibular incisors of Vertucci's type III ([Vertucci, 1984](#)) were matched.

2.2. Root canal preparation

We performed a standard access cavity using diamond burs 1012, 3082, and Endo-Z (Dentsply Sirona), and then a lingual wear to facilitate access to the lingual canal. The apical patency was confirmed by using a size 10 K-file (Dentsply Sirona) in the buccal canal, and then the instrument was precurved to be inserted into the root canal and directed to the lingual canal. The working length (WL) was established as 1 mm shorter from the apical foramen by introducing the file until its tip was visualized through the apical foramen. A glide path was performed with a size 15 K-file (Dentsply Sirona).

The teeth were randomly divided into 3 groups ($n = 12$) and prepared by inserting the instruments initially into the buccal canal and then precurving the instruments to direct them towards the lingual canal:

- HCM Group: the Hyflex CM (Coltene-Whaledent, Allstatten, Switzerland) size 25/0.08 instrument was used for the cervical preparation, and then the canals were instrumented up to the WL with in-and-out movements according to the following sequence of instruments sizes: 15/0.04 (300 rpm speed and 1.8 Ncm torque), 20/0.04, 25/0.04, 30/0.04, and 40/0.04 (500 rpm speed and 2.5 Ncm torque).

- HEDM Group: the Hyflex EDM (Coltene-Whaledent, Allstatten, Switzerland) instruments were inserted into the root canals with in-and-out movements using 500 rpm speed and 2.5 Ncm torque. The size 25/0.08 HCM instrument was used to prepare the cervical third. The 25/~ instrument was used until the WL was reached, and then the 40/0.04 instrument was used in the same manner.

- SRF Group: the Sequence Rotary File (MK-Life, Porto Alegre, RS, Brazil) instrument size 25/0.06 was used for the cervical third preparation, then the instruments were used in sequence with in-and-out movements until the WL was reached: 15/0.04 (300 rpm speed and 1.8 Ncm torque), 20/0.06, 25/0.06, 35/0.04, and 40/0.04 (400 rpm speed and 2.0 Ncm torque).

For all groups, the X-Smart Plus motor was used to power the instruments, and the root canals were irrigated with 3 mL of 1 % sodium hypochlorite using a 29-G needle (NaviTip, Ultradent Inc., South Jordan, UT, USA) after each instrument was used.

After preparation with 0.25 mm and 0.40 mm tip diameters instruments, passive ultrasonic irrigation (PUI) was performed using the E1-Irrisonic 20.01 ultrasonic point (Helse, Santa Rosa do Viterbo, SP, Brazil) coupled to the ultrasound device (NSK Varios 350) in Endo function with a power of 20 %, by introducing it into the root canal 2 mm from the WL with 3 mL of 1 % sodium hypochlorite and activating it for 20 s. The irrigating solution was then renewed and the activation cycle was repeated twice, totaling 60 s of ultrasonic agitation.

Each HCM, HEDM, and SRF instrument was used for the instrumentation of four specimens and then discarded. All preparations were performed by a single endodontist, trained and calibrated in the use of the tested instruments, who was also blinded to the 3D virtual models of the specimens. During root canal preparation, the samples were kept in saline solution at room temperature. There was no instrument fracture or sample loss, and no procedural error was observed.

2.3. Scanning and evaluation

The teeth were scanned after preparation with 0.25 mm and 0.40 mm tip diameters instruments using the micro-CT SkyScan 1174. The parameters used were the same as those used in the sample selection.

Once the instrumentation was concluded, both two-dimensional and three-dimensional images were registered with their respective reference images (scanned at sample selection) and target images (scanned after instrumentation) in the DataViewer software (versions 1.5.1, Bruker-microCT), and superimposed with geometric alignment for further analysis of the root canal.

2.4. Parameters

The images were analyzed using CTAn software (CTAn v.1.12, Bruker-microCT). The volume (mm^3) of the canal was measured in three segments: 1–4 mm (apical third), 4–7 mm (middle third), and 7–10 mm (cervical third) from the apical foramen. Dentin thickness was measured using the images taken before and after instrumentation to obtain the percentage of changes after the procedure.

Root canal transportation and centering ability were measured using the equation described by [Gambill et al. \(1996\)](#). The shortest distance between the mesial margin of the canal and the root (X) and the shortest distance between the distal margin of the canal and the root (Y), before (1) and after instrumentation (2), were calculated in millimeters. Each third was divided into 5 equal slices for measurement and the arithmetic mean of each third was used. In the root canal transportation equation, given as $(X1-X2) - (Y1-Y2)$, the 0 result means no canal transportation

and other values represent the direction of transportation (transportation to X for negative values and to Y for positive values). In the centering ability equation, given as $(X1-X2)/(Y1-Y2)$, the result of 1 indicates complete centering, and the closer the value to 0, the lower the centering. The same method was used to measure the transportation and centering ability of the instruments in the bucco-lingual direction, with X being the shortest distance between the buccal canal margin and the root and Y the shortest distance between the lingual margin of the canal and the root canal. All evaluations were performed by a single researcher calibrated to use the software.

When contrasting arithmetic and logical operations to the images superimposed on the CTAn Software, the percentage of debris (% debris) accumulated in the root canals and the percentage of untouched canal areas (% non-instrumented area) were calculated after instrumentation (Pinto et al., 2019).

2.5. Statistical analysis

Data were analyzed using the D’Agostino and Pearson tests to verify normality. ANOVA and Tukey tests were used for parametric data and Kruskal-Wallis and Dunn for non-parametric data. Statistical analysis was performed using the GraphPad Prism 8 software (La Jolla, CA, United States).

3. Results

The increase in volume with the increase in the instrument diameter tip from 0.25 mm to 0.40 mm was significant ($P < 0.05$) in the apical region for the HCM and SRF systems (Fig. 1). After preparation with the 0.40 mm tip diameter instrument, the SRF system provided a reduction in the buccal dentin thickness at 1 mm from the apex ($P < 0.05$) (Fig. 2). There was greater transportation in the bucco-lingual direction after both preparations (Tables 1 and 2). The systems were evaluated in the 0.25 mm and 0.40 mm diameters by thirds, and they did not show differences ($P > 0.05$) regarding untouched canal areas and accumulated debris (Table 3).

4. Discussion

In the primary anatomical study carried out for the selection of samples, it was detected that this particular root canal configuration, in which a single canal bifurcates and then merges again, may consist of a double curvature in the bucco-lingual direction that could interfere with the endodontic instrumentation. Therefore, Nickel-Titanium instruments with different heat treatments and flexibility were selected for sample preparation.

In the apical and middle thirds, after preparation with 0.25 mm tip diameter instruments, the SRF system provided a significant increase in root canal volume when compared to the HCM system, probably due to

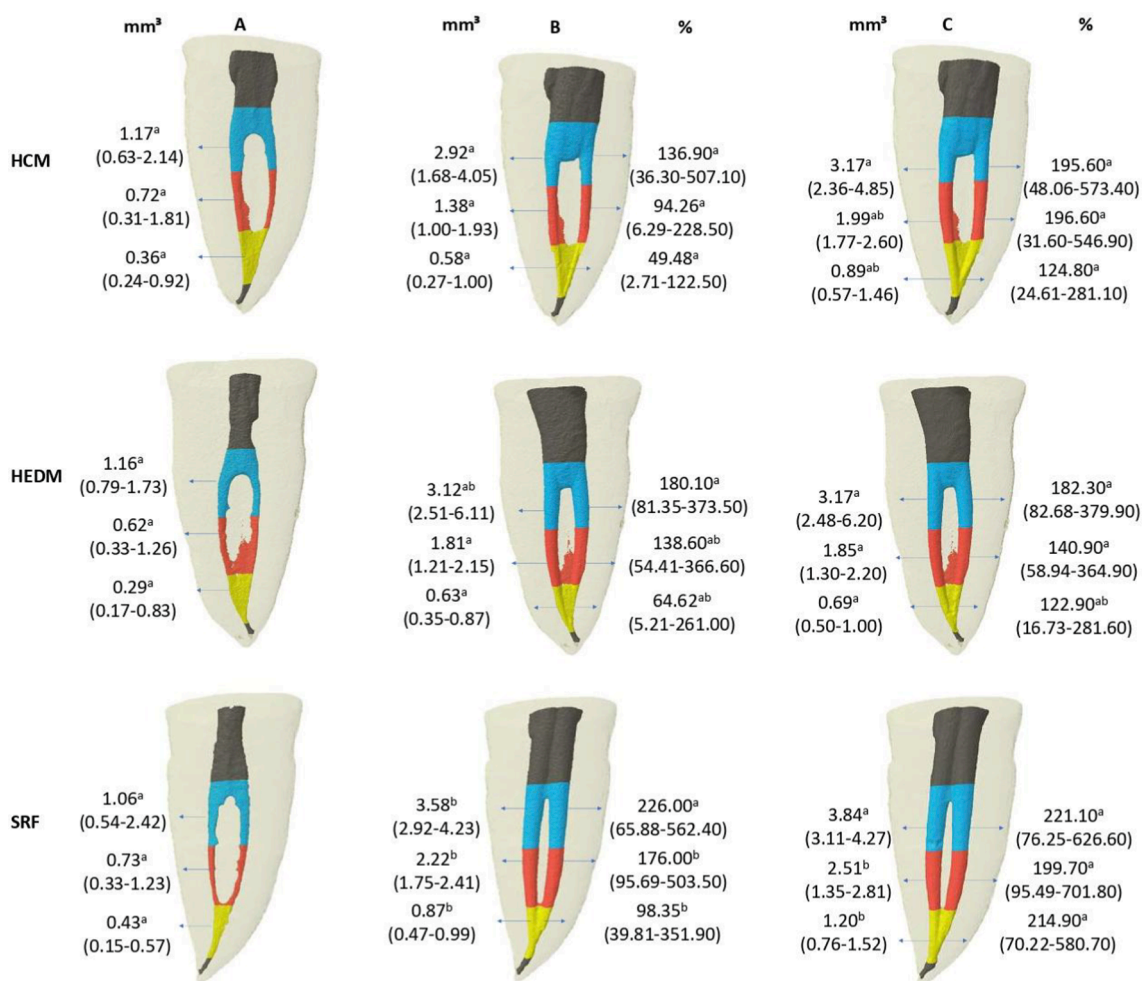


Fig. 1. Median, maximum, and minimum values of the original root canal volume (column A) and after preparation with tip diameter instruments of 0.25 (column B) mm and 0.40 mm (column C), volume increase in cubic millimeters (mm³) and percentage (%). In the image, the yellow, red, and blue colors represent the apical, middle, and cervical thirds of the root canal, respectively. Different superscript letters in each line represent a statistically significant difference between the groups. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

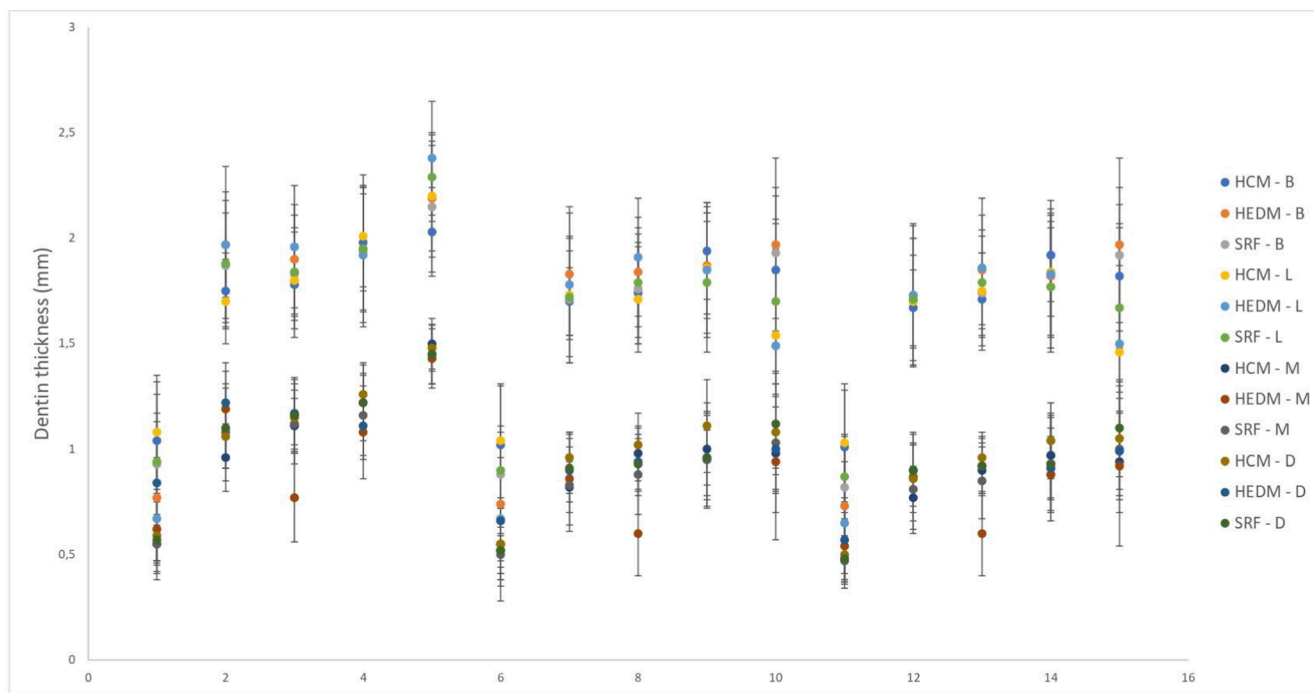


Fig. 2. Mean and standard deviation of the original dentin thickness and after apical preparation with tip diameter instruments of 0.25 mm and 0.40 mm. Thickness reduction of the buccal, lingual, mesial, and distal walls are represented in millimeters (mm).

Table 1

Median, maximum, and minimum values of transportation and mean and standard deviation of the centering ability in millimeters (mm) in the mesio-distal direction after apical preparation with 0.25 mm and 0.40 mm tip diameter instruments.

		Transportation		Center ability	
		0.25	0.40	0.25	0.40
H CM	Cervical	0.03 (-0.16 - 0.44) ^a	0.03 (-0.16 - 0.42) ^a	0.71 ± 0.18 ^a	0.72 ± 0.17 ^b
	Middle	0.01 (-0.04 - 0.06) ^a	0.005(-0.05 - 0.06) ^b	0.83 ± 0.09 ^a	0.85 ± 0.08 ^a
	Apical	-0.001 (-0.01 - 0.04) ^a	0.01 (-0.04 - 0.03) ^a	0.82 ± 0.11 ^a	0.86 ± 0.11 ^b
H EDM	Cervical	0.05 (-0.14 - 0.31) ^a	0.05 (-0.14 - 0.29) ^a	0.73 ± 0.17 ^a	0.74 ± 0.15 ^a
	Middle	0.03 (-0.02 - 0.10) ^a	0.04 (-0.02 - 0.11) ^b	0.82 ± 0.11 ^a	0.82 ± 0.12 ^a
	Apical	0.009 (-0.04 - 0.08) ^a	0.01 (-0.03 - 0.07) ^a	0.76 ± 0.13 ^a	0.82 ± 0.10 ^a
SRF	Cervical	-0.02 (-0.98 - 0.11) ^a	-0.02 (-0.38 - 0.14) ^a	0.71 ± 0.19 ^a	0.72 ± 0.19 ^b
	Middle	0.02 (-0.03 - 0.08) ^a	0.03 (-0.03 - 0.05) ^a	0.85 ± 0.09 ^a	0.86 ± 0.07 ^a
	Apical	0.001 (-0.03 - 0.03) ^a	-0.01 (-0.03 - 0.02) ^a	0.82 ± 0.13 ^a	0.89 ± 0.11 ^b

Different superscripted lowercase letters in each line indicate a significant difference between groups (P < 0.05). Negative results mean that the preparation was transported to X (mesial) direction and positive results mean that the preparation was transported to Y (distal) direction. The value of 1 means a centered canal, and the closer the value of 1, the more centered the preparation.

the taper of the instruments. The increase was from 93.54 % to 166.78 % after preparation with the 0.25 mm tip diameter instrument, and from 148.7 % to 211.9 % with the 0.40 mm tip diameter instrument. Such values are higher than those reported by Pérez (2018), 48 % and 102 % for 0.25 and 0.40 apical enlargement, respectively, and by Velozo et al. (2020), 107.50 % and 93.13 % for 0.30 and 0.40 apical enlargement, respectively. However, both studies were carried out in mandibular incisors with a single root canal, classified as Vertucci's type I.

In general, there was a significant reduction in the dentin thickness in the mesio-distal direction. Still, what must be considered is the dentin remnant. Elayouti et al. (2008) reported that a dentin wall remnant of up to 0.5 mm would represent a removal of more than 50 % in the constricted region and 75 % in the wide region of oval canals, and a measure smaller than this would represent a thin wall. Thus, null hypothesis number 1 was rejected, as a smaller mean remnant was observed after preparation with the 0.40 mm tip diameter instrument 1 mm from the

apex in the SRF and in the mesial wall with the HCM, which may be considered critical. All samples were anatomically matched and prepared with instruments with the same tip diameter and taper, 40/0.04, thus, the differences observed between the groups can be explained by the metallic alloy and cross-section design of the systems, which seems to have influenced the dentin cutting. The heat treatments of the systems used increase resistance and flexibility. The HCM system has an alloy with memory control, the SRF with blue treatment (a visible layer of titanium oxide resulting from post-machining heat treatment), and the HEDM is made of a memory alloy controlled using electrical discharge machining technology, which presents greater cyclic fatigue when compared to HCM and other blue treated systems (Das et al., 2019; Gündoğar and Özyürek, 2017). The reduction in thickness was less than 17 %, and in the other segments and systems evaluated, the preparation can be considered safe as there was a dentin thickness greater than 0.5 mm left.

Table 2

Median, maximum, and minimum values of transportation and mean and standard deviation of the centering ability in millimeters (mm) in the bucco-lingual direction after preparation with 0.25 mm and 0.40 mm tip diameter instruments.

		Transportation		Centering ability	
		0.25	0.40	0.25	0.40
H CM	Buccal	-0.02 (-0.11 - 0.04) ^b	-0.02 (-0.14 - 0.04) ^a	0.51 ± 0.20 ^a	0.50 ± 0.18 ^a
	Lingual	0.06 (-0.03 - 0.27) ^a	0.08 (-0.02 - 0.36) ^a	0.35 ± 0.31 ^a	0.44 ± 0.33 ^a
H EDM	Buccal	-0.03 (-0.26 - 0.02) ^{a,b}	-0.04 (-0.37 - 0.02) ^a	0.58 ± 0.37 ^a	0.49 ± 0.13 ^a
	Lingual	0.11 (-0.40 - 0.36) ^a	0.16 (-0.38 - 0.35) ^a	0.28 ± 0.18 ^a	0.30 ± 0.16 ^a
SRF	Buccal	-0.15 (-0.38 - 0.06) ^a	-0.14 (-0.35 - 0.27) ^a	0.33 ± 0.32 ^a	0.46 ± 0.38 ^a
	Lingual	0.19 (-0.11 - 0.70) ^a	0.19 (-0.02 - 0.30) ^a	0.32 ± 0.26 ^a	0.47 ± 0.50 ^a

Different superscripted lowercase letters in each line indicate a significant difference between groups (P < 0.05). Negative results mean that the preparation was transported to X (buccal) direction and positive results mean that the preparation was transported to Y (lingual) direction. The value of 1 means a centered canal, and the closer the value of 1, the more centered the preparation.

TABLE 3

Median, maximum, and minimum values of the percentage of untouched canal areas in millimeters (mm) and accumulated debris (in percentage) after preparation with 0.25 mm and 40 mm tip diameter instruments.

		Untouched	canal areas	Percentage of	debris
		0.25	0.40	0.25	0.40
H CM	Cervical	6.57 (0.32–14.49) ^{ab}	6.85 (0.32–18.70) ^{ab}	0.20 (0.01–0.46) ^{ab}	0.20 (0.00–0.82) ^{ab}
	Middle	20.73 (7.49–39.71) ^{aA}	19.74 (4.94–34.18) ^{bA}	2.07 (0.35–6.34) ^{aA}	1.85 (0.30–5.27) ^{aA}
	Apical	23.81 (2.51–79.33) ^{aA}	14.04 (0.74–51.42) ^{bA}	5.99 (0.84–23.33) ^{aA}	3.33 (0.27–15.44) ^{aA}
H EDM	Cervical	4.76 (3.05–12.60) ^{ab}	4.53 (0.13–9.15) ^{aA}	0.15 (0.01–1.00) ^{ab}	0.12 (0.00–0.53) ^{ab}
	Middle	10.30 (3.59–28.56) ^{aAB}	8.79 (4.09–35.76) ^{aAB}	1.87 (0.16–5.79) ^{aA}	1.99 (0.19–4.73) ^{aA}
	Apical	19.05 (2.071–59.79) ^{aA}	20.80 (1.11–58.38) ^{aA}	3.43 (0.07–18.28) ^{aA}	2.70 (0.06–18.50) ^{aA}
SRF	Cervical	6.00 (1.84–15.44) ^{ab}	4.97 (0.40–13.36) ^{bb}	0.13 (0.01–0.50) ^{ab}	0.17 (0.00–0.49) ^{ab}
	Middle	13.40 (6.18–24.45) ^{aAB}	10.62 (4.20–19.92) ^{bAB}	1.32 (0.34–4.95) ^{aA}	1.01 (0.01–3.71) ^{bA}
	Apical	15.69 (1.53–36.01) ^{aA}	11.54 (1.35–26.09) ^{bA}	1.97 (0.03–6.73) ^{aA}	1.48 (0.13–4.46) ^{aA}

Different superscripted lowercase letters in each line indicate a statistically significant difference between preparations up to the apical diameter of 0.25 mm and 0.40 mm. Different superscripted capital letters in each column indicate a statistically significant difference between the thirds.

The transportation of the root canal in the bucco-lingual direction was greater after the 0.25 mm and 0.40 mm preparations in all groups evaluated, especially on the lingual wall. These preparations cannot be considered centralized. This greater transportation can be explained by the internal anatomical configuration of these teeth, as the root canal bifurcates and then merges again. Consequently, the internal anatomy of these teeth ends up forming a double curvature in bucco-lingual direction, causing the instrument to enter in a straightforward movement towards the buccal or lingual directions and then, when the canal is unified, it becomes straight again, causing two points of tension and fatigue in the instrument, which justifies the use of more flexible instruments. Despite the instruments' flexibility, this bifurcation causes them to be forced against these walls, especially the lingual one, since access to this canal is more difficult. Different factors can influence canal transportation and centering ability, including the instrument design (cross-section, taper, tip), which can provide greater cutting power and, consequently, more canal transportation (Sousa-Neto et al., 2018). There is no current data available in the literature regarding the transportation and centering ability in this dental group for comparison.

The second null hypothesis can be accepted because there was no statistically significant difference among the three systems concerning the percentage of untouched canal areas. In all of them, the apical third presented a higher percentage of untouched canal areas, contrary to what Azim et. al (2017) observed, with the apical third having the lowest percentage. This may be because they provided greater apical enlargement of single canals, where there is no bifurcation of the root canal. This facilitates access to the apical region, where the instrument can reach all the areas. The percentage values of untouched canal areas in the apical third after preparation with 0.25 mm tip diameter

instruments were lower than those reported by Pérez et al. (2018) after the preparation of Vertucci's type I root canals with 0.25 mm and 0.30 mm tip diameter instruments (30.3 %). The values observed in the groups after 0.40 mm preparation were close to those observed by Velozo et al. (2020) (13.08 % and 11.74 %), but smaller in the apical third for the HCM and SRF systems after preparation with 0.40 mm tip diameter instruments when compared to 0.35 mm or 0.40 mm tip diameter instruments in the study by Pérez et al. (2018) (15.9 %), and lower for all systems when evaluating the total percentage of untouched canal areas (20.1 %).

A considerable percentage of debris in the root canal was detected after instrumentation. The apical third showed a greater accumulation, in disagreement with the literature (Gavini et al., 2018). The main reason is the presence of a bifurcation in the root canal, which increases the difficulty of access to the apical third — especially for the irrigation needles — and hinders an adequate flow of the irrigating solution and removal of debris. On the other hand, the cervical third had a lower accumulation due to the ease of access and better flow of the irrigating solution, facilitating the removal of debris, as observed by Azim et al. (2017).

5. Conclusion

The preparation with HCM, HEDM, and SRF instruments with a tip diameter of 0.40 mm proved to be effective, leaving a safe dentin remnant, showing no significant differences regarding the transportation and centralization of the root canal, and reducing the untouched canal areas, mainly in the apical third, for all systems.

Ethics approval

All procedures performed in studies involving human participants were accordance with the ethical standards of the local Institute Review Board (School of Dentistry of Bauru, University of São Paulo (FOB/USP), Bauru, São Paulo, Brazil) (#1.051.377).

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CRedit authorship contribution statement

Renata Maira de Souza Leal: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Resources, Software, Visualization, Writing – original draft. **Felipe Andretta Copelli:** Data curation, Methodology, Writing – review & editing. **Jáder Camilo Pinto:** Formal analysis, Resources, Writing – review & editing. **Mario Tanomaru-Filho:** Methodology, Validation, Writing – review & editing. **Marco Antonio Hungaro Duarte:** Methodology, Supervision, Validation, Writing – review & editing. **Bruno Cavalini Cavenago:** Conceptualization, Funding acquisition, Investigation, Methodology, Project administration, Supervision, Validation, Visualization, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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