Article published online: 2021-11-01

Original Article

Micro-CT Analysis of Two-Dimensional and Three-Dimensional Parameters in Severely Curved Roots Prepared with 3 Instrumentation Systems: An *In vitro* Study

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Abstract

Aim: This study analyzed the two-dimensional (2D) and three-dimensional (3D) parameters and canal transportation using micro-CT in roots prepared with three instrumentation systems preceded or not by glide path. **Materials and Methods:** Sixty mesial canals of mandibular molars with severe curvature angles (≥25°) were divided randomly ($n = 10$): G1-Twisted-file-adaptive (TFA); G2-Reciproc (RC); G3-One-Shape (OS); G4-ProGlider (PG)+TFA; G5-PG+RC; and G6-PG+OS. Area, perimeter, circularity, and major and minor diameters at the apical 5 mm and volume and surface area and canal transportation were evaluated. Data were analyzed using two-way analysis and Tukey's test ($\alpha = 0.05$). **Results:** RC showed significantly higher areas and lower perimeters and diameters than TFA ($P < 0.001$). Increases in root canal volumes promoted by RC and OS were similar ($P = 0.979$). There was no statistically significant difference in canal transportation ($P = 0.083$). RC generally promoted major changes in the 2D and 3D parameters. **Conclusions:** The RC system caused a significantly higher perimeter and area increase than the TFA system. The preceding use of PG in the instrumentation techniques did not influence the evaluated parameters.

Keywords: Endodontics, instrumentation systems, micro-CT scan, root canal preparation, severely curved roots

Introduction

The instrumentation of the curved root canals presents a challenge considering the definite risks of canal transportation and perforation.[1,2] In addition, in curved roots, the instrument cannot promote adequate three-dimensional (3D) preparation, which leaves a significant percentage of the canal surface area untouched.[3] The permanence of debris may predispose patients to persistent infection and consequently compromise the root canal treatment.[4]

To minimize the limitations imposed by this anatomical complexity, instruments have undergone modifications in the composition of their alloys to improve their flexibility. Reciproc (RC) instruments (VDW, Munich, Germany) are made of an M-Wire nickel-titanium (NiTi) alloy, which has greater flexibility and cyclic fatigue resistance than traditional NiTi alloys.[5] However, the pressure exerted on the instrument during its use increases the possibility of canal transportation.[6] The One-Shape (OS) (OS; Micro Mega, Besancon, France) is composed of a conventional superelastic austenite 55-NiTi alloy.[7] The files have variable pitch length along the entire blade, which limits the risk of minimal instrument fatigue, consequently eliminating the risk of instrument breakage.[8]

The twisted-file-adaptive (TFA) system (SybronEndo, Orange, CA, USA) consists of a sequence of three NiTi instruments for use in a specific reciprocating motion.[9] These instruments are created by the transformation of a raw NiTi wire in the austenite crystalline structure phase into a different phase

Access this article online	
Quick Response Code:	**Website:** www.ejgd.org
	DOI: 10.4103/ejgd.ejgd_140_19

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How to cite this article: Souza-Flamini LE, Crozeta BM, Silva RG, Savioli RN, Sousa-Neto MD, Cruz-Filho AM. Micro-CT analysis of two-dimensional and three-dimensional parameters in severely curved roots prepared with 3 instrumentation systems: An *in vitro* study. Eur J Gen Dent 2020;9:62-8.

Submitted: 02-Sep-2019 **Accepted:** 07-Jan-2020 **Published:** 29-Apr-2020

of crystalline structure (R-phase). Such a treatment confers instrument flexibility, allowing it to adjust the torque values such that the elastic limit is not exceeded.[10]

The literature shows that TFA provides better root canal centralization than do systems that work with continuous and alternate rotational kinematics.[11,12] Another study suggested that rotary glide path preparation before biomechanical performance improves the ability to shape the area while following the original root canal curvature.[13] The ProGlider (PG) (Dentsply Sirona, Ballaigues, Switzerland) is a rotary glide path instrument manufactured from the M-Wire alloy. The PG has a tip diameter of 0.16 mm and a progressive taper ranging from 2% to 8%.[14]

Micro–computed tomography (micro-CT) has been used to evaluate the morphological changes in the root canal generated by biomechanics since it offers more detailed images and sample preservation.[15]

Morphological alterations occurring in dentin during excision are generally beneficial for sanification, as long as they do not promote canal transportation. Therefore, this study analyzed the two-dimensional (2D) and 3D parameters and canal transportation of severely curved roots prepared by three mechanical systems with different kinematics preceded or not by glide path using micro-CT. The null hypothesis was that there would be no difference in the 2D and 3D parameters between systems with or without glide path preparation.

Materials and Methods

Sample selection and preparation

The study was approved by the institute's research ethics committee (CEP/FORP-USP, N. 2010.1.1478.58.5). A total of 170 human mandibular molars with complete root and apical formation were selected from the human permanent tooth bank of the School of Dentistry of Ribeirão Preto.

The teeth were radiographed with a digital radiograph sensor (IDA; Dabi Atlante, Ribeirão Preto, SP, Brazil). The exclusion criteria were pulp nodules, root canal and/or pulp chamber calcification, internal resorption, previous root canal treatment, and perforated root. A total of 151 maxillary molars remained after the preselection. The same radiographic images were used to determine the curvature angles and radius. The Dimension Angle tool of CorelDraw Graphics Suite X6 software (Corel, Ottawa, Canada) was used for this purpose. The angle severity pattern followed the recommendations of Schneider (1971). Thus, a line (a) was drawn from the entrance of the root canal, following the long axis of the root canal. Concomitantly, a second line (b) was drawn, extending from the end of the apical foramen to the intersection with the first line. The point of intersection of the lines indicated the beginning of the curvature [Figure 1]. Molars that exhibited mesial roots with an angle of curvature ≥25° were selected. Subsequently, the radius of curvature was determined following the recommendations of Pruett *et al.* (1997). Thus,

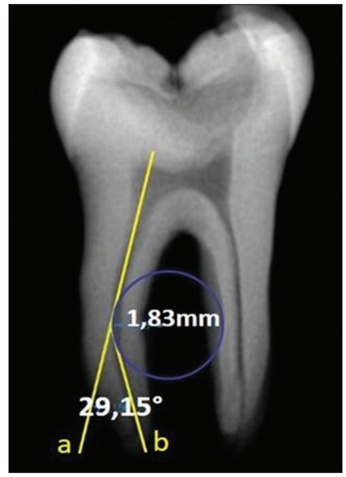

Figure 1: Determination of root canal curvature and radius of curvature

we identified the point at which the root canal begins to deviate from its long axis in each of the lines drawn (a, b), to determine the angle of curvature. Finally, a circle encompassing the two previously defined points was drawn. The radius of the circle was defined as the radius of curvature [Figure 1].

Among them, 105 molars with severe curvature (≥25°) and radius of curvature (≤5 mm) were selected. The specimens were mounted in a sample holder and subjected to micro-CT scanning (SkyScan 1174 v2; Bruker-microCT, Kontich, Belgium). The parameters used were 50 kV, 800 mA, 360° of rotation with a step size of 1°, and an isotropic resolution of 22.90 μm. Of the 105 scanned specimens, the first 30 teeth that had two independent root canals with two separate apical foramina in the mesial root were selected.

Images of all specimens were reconstructed using NRecon v.1.6.3 software (Bruker-microCT, Kontich, Belgium), which provided axial cross-sections of the inner structure of the roots in bitmap format. Morphological 2D (area, perimeter, circularity, and major and minor diameters) and in 3D parameters (volume and surface area) of the root canal were investigated using CTAn v.1.14.4 software (Bruker-microCT, Kontich, Belgium).

Root canal preparation

The experimental groups consisting of 60 root canals of 30 molars were distributed using a stratified sampling technique. Six groups (n = 10) were formed and paired by length, volume, and area of each root canal. The sample data were normally distributed (Shapiro–Wilk, $P > 0.05$) with homogeneous variance (Levene's test, $P > 0.05$), confirming the uniformity of selection.

After endodontic access was achieved, the working length (WL) of each root canal was determined by subtracting 1 mm from the length where size 10 was visible at the apical foramen. Biomechanical preparation was performed according to the manufacturer's recommendation: Group 1, TFA instruments (size 20, 0.04 taper; size 25, 0.06 taper). TFA size 20.04, followed by a 25.06 instrument were taken into the root canal until WL was achieved and powered by an electric Sybron Elements motor (SybronEndo, Orange, CA, USA); Group 2, RC (size 25, 0.08 taper). The instrument R25 was operated in a reciprocating motion by an electric VDW Silver motor (VDW, Munich, Germany). The instrument was introduced into the root canal until resistance was felt and then activated in an apical direction using in-and-out pecking motion about 3 mm in amplitude with light apical pressure. After the three pecking motions were made, the instrument was removed from the root canal and cleaned; Group 3, OS instrument (size 25, 0.06 taper). The OS instrument was used with a rotary motion by an electric X-Smart Plus motor (Dentsply Sirona, Baillaigues, Switzerland) to reach 2/3 of the WL, WL – 3 mm, and the WL; Group 4, glide path with a PG instrument followed by TFA instruments; Group 5, glide path with a PG instrument followed by a RC; and Group 6, glide path with a PG instrument followed by an OS instrument. Initially, in these groups, the PG instrument was used in one or more passes until the WL was achieved, and then the different systems were used. All of the instruments were operated using a torque-controlled endodontic motor according to the manufacturers' recommendations. Thus, the TFA instrument was operated in the TF Adaptive program Elements motor (SybronEndo, Orange, CA, USA), the RC was operated in the "Reciproc ALL" program (VDW, Munich, Germany), the OS was operated at 400 rpm and 2.5 Ncm torque, and the PG was operated at 300 rpm and 2 Ncm torque. All instruments were scanned and discarded after each use. During preparation, 3 mL of a 1% sodium hypochlorite solution was used between each file or after three pecking motions. In all groups, the biomechanical procedure was performed by a specialist endodontist (Souza-Flamini L.E.), who had clinical experience with the use of the three systems assessed.

The prepared roots were subjected to micro-CT scanning (SkyScan 1174 v2; Bruker-microCT, Kontich, Belgium). The pre- and post-preparation images [Figure 2] were analyzed using the CTAn v. 1.14.4.1+ (Bruker-microCT, Kontich, Belgium) to calculate canal transportation.

Statistical analysis

Two-way analysis of variance was used to evaluate the influence of the glide path (with and without glide) and file instruments (TFA, RC, and OS), followed by Tukey's test ($\alpha = 0.05$) using SigmaPlot v11.0 (Systat Software, Chicago, IL, USA). The confidence level was 95%.

Results

Two-dimensional analysis

Table 1 shows the 2D parameter analysis. Without the glide path, the increase in the root canal area promoted by the RC in the final apical 5-mm section was significantly higher than that promoted by the TFA ($P < 0.001$). No significant difference was observed between OS and the other techniques ($P = 0.744$). With the glide path, the increase in the root canal area promoted by the RC was significantly higher than that promoted by the OS and TFA instruments ($P < 0.001$). No significant difference

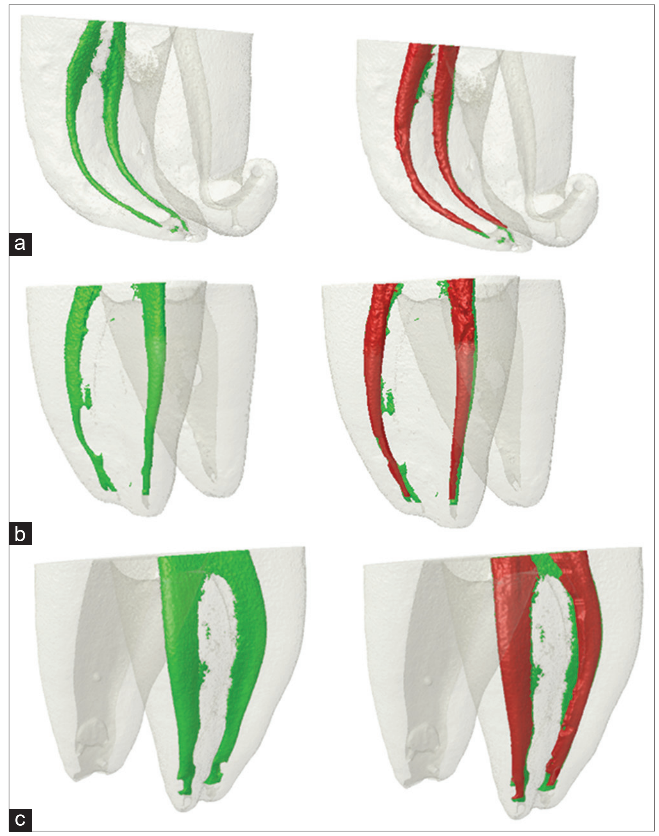

Figure 2: Three-dimensional models of root canal systems of the mesial root of mandibular molars before and after biomechanical preparation with the Twisted-File Adaptive (a), Reciproc (b), and One-Shape (c) systems. The green color indicates uninstrumented areas of the canals, whereas the red color indicates instrumented areas

was detected between the latter two instruments ($P = 0.274$). The instrumentation with or without the glide path did not have any influence on increasing the root canal area regardless of the system used ($P = 0.744$). The mean percentage of perimeter increase in the root canal was higher for RC, followed by TFA and OS ($P < 0.001$). The use of PG did not influence the perimeter increase compared with that of the system alone ($P = 0.620$). The higher mean percentage of diameter increase in the final 5-mm section of the root canal was provided by RC, followed by TFA, and there were significant statistical differences between them ($P < 0.001$). The increase in the minor canal diameter was not influenced by the use of PG ($P = 0.812$).

Three-dimensional analysis

The 3D parameter analysis is shown in Table 2. There was no significant difference in the root canal final volumes promoted by RC and OS ($P = 0.409$). However, both instruments promoted a significant increase in root canal volume compared with TFA ($P < 0.001$) regardless of the use of PG ($P = 0544$). The increased surface area obtained with RC was significantly higher than that obtained with TFA ($P = 0.024$). There was no significant difference between OS and the other instruments. The previous use of PG instrumentation did not affect the root canal surface area ($P = 0.123$).

Table 1: Analyze the two-dimensional parameters (means of the final 5 mm)

	Without PG			With PG		
	TFA	RC	OS	TFA	RC	OS
Area						
Original	0.12±0.07	0.13±0.05	0.13±0.05	0.12±0.04	0.11±0.04	0.13±0.06
Δ	0.07±0.03B	0.13±0.04A	0.10±0.07AB	0.08±0.05b	0.13±0.06a	0.08±0.07b
Δ (%)	78.49±51.24	148.0±114.0	85.77±67.59	87.67±74.38	139.28±89.25	84.59±92.66
Perimeter						
Original	1.26±0.23	1.31±0.29	1.30±0.16	1.20±0.31	1.28±0.19	1.27±0.18
Δ	0.40±0.40B	0.55±0.24A	0.25±0.21C	0.39±0.23b	0.50±0.22a	0.27±0.20c
Δ (%)	34.26±31.63	46.30±28.96	22.73±22.72	34.74±23.92	39.43±21.77	23.42±21.37
Roundness						
Original	0.52±0.19	0.60±0.17	0.44±0.21	0.53±0.18	0.59±0.17	0.44±0.19
Δ	0.17±0.19A	0.24±0.19A	0.22±0.25A	0.17±0.22a	0.26±0.18a	0.24±0.23a
Δ (%)	47.08±60.89	56.16±62.17	91.34±126.99	49.10±72.26	51.94±62.63	76.14±82.04
Major diameter						
Original	0.47±0.14	0.48±0.14	0.50±0.14	0.44±0.13	0.46±0.13	0.50±0.14
Δ	0.11±0.10A	0.12±0.11A	0.11±0.11A	0.12±0.15a	0.12±0.09a	0.09±0.08a
Δ (%)	25.95±22.77	32.70±34.80	26.15±28.98	27.52±30.89	28.89±2.72	21.38±23.57
Minor diameter						
Original	0.28±0.07	0.32±0.08	0.30±0.10	0.31±0.08	0.30±0.07	0.32±0.12
Δ	0.13±0.07C	0.22±0.08A	0.16±0.09B	0.10±0.08c	0.20±0.10a	0.15±0.10b
Δ (%)	55.21±39.38	79.65±43.21	62.93±44.47	36.62±28.88	73.70±47.17	60.35±50.41

Different uppercase letters in rows are designed to compare instrumentation systems without PG ($P<0.05$), Different lowercase letters in rows are designed to compare instrumentation systems with PG ($P<0.05$), Δ – Mean±standard deviation, TFA – Twisted-file-adaptive, OS – One-Shape, RC – Reciproc, PG – ProGlider

Table 2: Analyze the three-dimensional parameters

	Without PG			With PG		
	TFA	RC	OS	TFA	RC	OS
Volume						
Original	1.63±0.64	1.76±0.78	1.75,±0.81	1.63±0.49	1.70±0.67	1.84±0.89
Δ	1.07±0.36B	2.03±0.45A	2.01±0.48A	1.22±0.23b	1.85±0.53a	1.83±0.50a
Δ (%)	72.1±25.7	136.2±67.0	149.0±89.1	88.2±32.1	121.2±48.3	119.6±75.8
Surface area						
Original	17.84±5.10	20.62±5.62	22.84±7.70	20.20±4.26	20.08±4.77	23.75±7.12
Δ	3.40±1.31B	5.47±1.38A	4.94±1.84AB	3.49±1.14b	4.77±1.39a	3.91±1.11ab
Δ (%)	24.9±17.0	28.8±11.5	28.4±16.9	23.6±14.5	26.4±9.9	18.7±9.6

Different uppercase letters in rows are designed to compare instrumentation systems without PG ($P<0.05$), Different lowercase letters in rows are designed to compare instrumentation systems with PG ($P<0.05$). *In rows represent statistical significant difference between instrumentation systems without PG and with PG ($P<0.05$). Δ – Mean±standard deviation, TFA – Twisted-file-adaptive, OS – One shape, RC – Reciproc, PG – ProGlider

Root canal transportation

A significant statistical difference was observed in the values of canal transportation between the instrumentation systems in only the cervical third [$P = 0.045$; Table 3]. For this third, OS yielded a higher rate of canal transportation than did RC ($P < 0.001$). There was no significant difference between OS and TFA. PG use had no effect on canal transportation ($P = 0.124$).

Discussion

In the present study, 2D and 3D morphological changes, as well as canal transportation caused by the continuous motion (OS), one reciprocating single-file system (RC), and one combined continuous/reciprocation motion (TFA), were evaluated using micro-CT scanning. The results revealed that the systems assessed showed significant differences in the parameters studied. Thus, the null hypothesis was rejected.

Micro-CT scanning offers a simple and reproducible technique for a 3D noninvasive assessment of the root canal system, and it can be applied quantitatively as well as qualitatively.[11] This method has been used to assess the effects of instrument systems on root canal geometry and transportation.[12] Mesial roots of extracted mandibular molars were used in this study. Even though simulated root canals using prefabricated resin blocks allow better standardization of the sample, the hardness and abrasion of acrylic resin and root dentin are not identical,

Table 3: Mean values±standard deviation of root canal transportation for each root canal third

Root third	Without PG			With PG		
	TFA	RC	OS	TFA	RC	OS
Cervical	0.13±0.05AB	0.13±0.03B	0.17±0.07A	0.12±0.05ab	0.18±0.05b	0.15±0.04a
Middle	0.11±0.03A	0.11±0.05A	0.15±0.07A	0.08±0.04a	0.12±0.07a	0.19±0.07a
Apical	0.15±0.08A	0.11±0.04A	0.14±0.06A	0.17±0.12a	0.10±0.04a	0.15±0.10a
Total	0.13±0.06AB	0.11±0.04B	0.15±0.07A	0.12±0.08ab	0.11±0.06b	0.14±0.08a

Different uppercase letters in rows are designed to compare instrumentation systems without PG ($P<0.05$), Different lowercase letters in rows are designed to compare instrumentation systems with PG ($P<0.05$). TFA – Twisted-file-adaptive, OS – One shape, RC – Reciproc, PG – ProGlider

and consequently does not reflect the action of the instruments in root canals of human teeth.[16,17]

The RC instrument caused higher increases in the area, perimeter, and minor diameter than did the other systems, regardless of the use of PG, confirming previous reports.[12,18] The increase in these parameters suggests a greater cutting capacity of the instrument.[12] Their cutting capacity results from a complex relationship among different parameters, such as the sectional design of the instrument, cutting angle of inclination, metallurgical properties, metal surface of active parts, and motion kinematics.[6] The TFA and OS systems have triangular cross-sectional shapes, whereas the RC system has variable tapers, a sharp double cutting edge, an S-shaped geometry, and a smaller cross-sectional area, with greater cutting capacity.[19] It should also be considered that although the instruments of the three tested systems had #25 diameter, the RC has the largest taper, consequently promotes greater divergence of the root canal walls.

The increased area and perimeter imply a greater enlargement of the root canal diameter. The removal of a greater amount of contaminated dentin is desirable in the case of necrotic teeth, as bacteria can penetrate dentinal tubules to a depth of approximately 420 µm.[20]

The three systems assessed were similar in relation to the circularity parameter. Considering that circularity values range from 0 to 1, where “0” corresponds to a straight line and “1” to a perfect circle; the mean values of 0.17–0.26 in the present study indicate that the root canal had a flattened shape at the end of root canal treatment. Thus, the preparation of the complete circumference of the root canal is probably not possible with any system. Instrumentation systems, using NiTi or stainless steel instruments, are not able to prepare 100% of the circumference of curved and flat root canals.[18]

Root canal volume is a variable used to analyze the effects of root canal instrumentation on dentin removal. In the present study, the data showed that RC had a significantly better volume and area increase than did TFA. The section design, taper angle, and cutting capacity may explain this result. The more refined cut associated with greater conicity resulted in a greater amount of excised dentin in more divergent root canal walls and a consequently greater root canal volume.[6,13] Notably, root canals that present an anatomical configuration with these parameters favor the irrigation or aspiration of the irrigating solution, making the filling phase easier. However, excessive root canal wear can promote root weakening or perforation. A similar result was reported in the literature.[13] A study points out that although RC removes a greater amount of dentin than do WaveOne and TFA, approximately 20%–35% of the surface of the root canal was found to have no instrumentation after preparation in the three groups.[13] Untouched dentin provides a reservoir for biofilms and allows persistent infection.[4]

One study reported that the glide path minimizes the risk of canal transportation.[16] The results showed that in the middle and apical thirds, canal transportation was similar to the RC, OS, and TFA systems, regardless of the use of PG. The lack of any influence of the glide path on canal transportation corroborates the findings of previous studies.[21-23] Flexibility may influence the instrument’s ability to properly shape curved root canals.[24] The RC and TFA instruments are composed of NiTi produced with M-Wire thermomechanical processing, which in the literature shows better properties in terms of flexibility and resistance to mechanical stress.[25] In contrast, the OS instrument is made of traditional NiTi alloys.[26] Based on the results of the present study, this thermal treatment does not appear to increase the instrument’s flexibility under our simulated clinical conditions. However, canal transportation is also related with instrument cross-section.[27] Different cross-sectional shapes can promote higher or lower flexibility of the instrument.[27] Therefore, a less flexible instrument may yield similar canal transportation performance results as reported by Brasil *et al.*[28] Another study[16] reported that the glide path affects the transportation of the root canal. These contrasting results can be explained by the type of instrument used. In this study, the glide path was created with three types of instruments: 1 made of stainless steel and 2 of NiTi. Canal transportation was significantly more frequent with the use of the stainless steel instrument, with no difference between the other two. The rigidity and lower flexibility of the stainless steel instrument compared to the NiTi instruments favored canal transportation. Although the results of the present study indicate that the creation of the glide path does not interfere with transportation of the root canal, previous work emphasizes that the glide path decreases the working time.[16,23,29] as well as extrusion of material beyond the apical foramen.[16] The extrusion of debris may cause postoperative complications and pain.[26] Thus, to obtain safe and efficient outcomes, the establishment of a glide path before root canal treatment is still recommended. In this study, the morphological changes of the root canal with the use of the three systems were assessed in the apical 5 mm of the root canal. The apical third is the critical area of the root canal, and remaining pulpal and inorganic debris have been detected in this area.[30] Thus, based on the results obtained and considering the limitation of an "*ex vivo*" study, CR was more efficient in promoting enlargement of the apical third without leading to significant canal transportation compared with the other systems. This observation may have clinical relevance mainly in cases of pulp necrosis where the removal of contaminated dentin is essential for the success of the treatment.

Conclusions

Use of the RC instrument produced greater modifications in most of the 2D and 3D parameters, which can be a positive factor mainly in the treatment of teeth with root canal infections. The RC, OS, and TFA systems were similar in terms of canal transportation, and although the glide path did not influence the parameters assessed, its clinical use should be taken into consideration.

Acknowledgment

The authors deny any conflicts of interest. We affirm that we have no financial affiliation (e.g., employment, direct payment, stock holdings, retainers, consultantships, patent licensing arrangements or honoraria), or involvement with any commercial organization with direct financial interest in the subject or materials discussed in this manuscript, nor have any such arrangements existed in the past three years.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

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