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# Shaping ability of Procodile and R6 Reziflow nickel-titanium reciprocating instruments in curved mesial root canals of mandibular molars: A MicroCT study

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Review Editor: Paolo Bianchini

## Abstract

To compare the shaping ability of Procodile and R6 Reziflow instruments used in reciprocating motion in severely curved root canals, assessed with micro-computed tomography ( $\mu$ CT). Fourteen extracted human mandibular first molars were randomly assigned to two instrumentation techniques ( $n = 14$  mesial root canals): Procodile or R6 Reziflow. For both groups, root canals were prepared to the working length up to a size 25, .06 taper. Molars were virtually divided into apical, middle and coronal thirds and  $\mu$ CT was used to scan all samples pre- and post-root canal. Canal transportation, centring ability, volume, surface area and unprepared area were evaluated. Geometrical parameter changes were compared with preoperative values (one-way analyses of variance and Tukey multiple comparison post-hoc test) between groups and Student *t*-test within groups ( $\alpha = 0.05$ ). Significantly less transportation was observed associated with the Procodile technique in the molar's coronal third compared to the R6 Reziflow technique ( $p < .05$ ). No significant differences in root canal centring ability, volume, surface area and unprepared area were observed. Procodile showed a lower percentage increase of surface area compared to R6 Reziflow ( $p < .05$ ). The Procodile and R6 Reziflow techniques applied to first molar root canal performed similarly except for the less transportation observed in the coronal third using Procodile.

## Research Highlights

MicroCT analysis of canal geometry before and after instrumentation revealed that Procodile and R6 Reziflow showed a similar shaping ability to shape curved root canals without substantially modifications of the original tooth anatomy.

## KEYWORDS

micro-CT, Procodile, R6 Reziflow, reciprocating, shaping ability

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## 1 | INTRODUCTION

Chemo-mechanical preparation of root canals aims to disinfect the root canal system while maintaining the original canal anatomy (Saber et al., 2017; Zavattini et al., 2020). Nickel-titanium (NiTi) endodontic instruments offer greater flexibility compared to stainless steel, thereby reducing procedural errors and operating time (Walia et al., 1988; Generali et al., 2014). Nevertheless, all instruments modify the original root canal anatomy, especially in curved canals, due to a nonuniform distribution of stress in contact points of the instrument and the canal (Peters, 2004; Zhang & Hu, 2010). Additional thermal treatments can significantly enhance the performances of rotary files, promoting the respect of the original canal anatomy (Yahata et al., 2009).

Procodile (Komet, Brasseler GmbH & Co., Lemgo, Germany) is a reciprocating instrument developed from the R6 Reziflow (Komet) instrument: the core area (tip to shaft) thickness is reduced, rendering the instrument more flexible for the optimum preparation of the curved root canals, and it has enlarged chip space, enabling superior dentin removal (Di Nardo et al., 2019; Generali et al., 2020). Conventional NiTi alloy, S cross section, taper (0.06) and tip diameters (#25) are the same as the R6 Reziflow. Procodile and Reziflow files revealed an almost austenitic phase structure at room and body temperature as confirmed by DSC and XRD analyses (Generali et al., 2020).

Furthermore, Procodile files can be operated with either traditional reciprocating endodontic motors or the dedicated motor EndoPilot (Komet, Brasseler GmbH & Co., Lemgo, Germany) which has the additional "ReFlex" movement (Komet. Procodile brochure, 2021).

Micro-computed tomography ( $\mu$ CT) is an accurate and non-destructive method to evaluate the effectiveness of NiTi endodontic instruments in terms of shaping ability and apical transportation (Drukteinis et al., 2019; Saber et al., 2017).  $\mu$ CT has been widely employed in dentistry and is considered the gold standard evaluation tool for geometrical changes within root canal following instrumentation.  $\mu$ CT produces high quality resolution images (around 20  $\mu$ m) (Peters et al., 2003).

Currently, there is no data available regarding the shaping ability of Procodile and R6 Reziflow instruments. Therefore, this study aims to evaluate the centring ability, canal transportation, canal volume, surface area and unprepared areas with the Procodile instruments compared to R6 Reziflow, as assessed with  $\mu$ CT. The null hypothesis assumed no differences between the two files in maintaining the original root canal anatomy.

## 2 | MATERIALS AND METHODS

Fourteen selected human mandibular first molars extracted for periodontal reasons, were used in this study. Criteria specified the inclusion of sound, completely formed teeth with the mesial root with a type II Vertucci's canal configuration (two separated mesial canals) (Vertucci, 2005) and with curvature angles between 20° and 45°, verified by standardized periapical radiographs (Schneider, 1971). Teeth with the presence of fractures, cracks, root caries or restorations were excluded. Ethical approval was obtained from Yorkshire & The

Humber-Leeds East Research Ethics Committee and registered with the National Health Service (NHS) England Research Authority (18/YH/0465).

Following a pilot study, a sample size of 7 teeth for each group was estimated 80% power (type-I error of 0.05), calculated with the G\*Power 3.1.9.2 software (Heinrich-Heine-Universität Düsseldorf, Düsseldorf, Germany).

All selected teeth were conserved in distilled water for up to 1 month at 4°C.

### 2.1 | Micro computed tomography scanning

Specimens were scanned using a Scanco uCT50 microCT scanner (Scanco, Brüttisellen, Switzerland). Samples were immobilized in 19 mm scanning tubes (using cotton gauze) and scanned to produce 20  $\mu$ m voxel size volumes, using X-ray settings of 90 kVp, 155  $\mu$ A and a 0.5 mm aluminum filter to attenuate harder X-rays. Scans were automatically scaled at reconstruction using the calibration object, provided by the CT manufacturer, consisting of five rods of hydroxyapatite (concentrations between 0 and 790 mg/cm<sup>3</sup>), with absorption values expressed in Hounsfield Units (HU). Specimens were characterized using Parallax Microview software package (Parallax Innovations Inc., Ilderton, ON Canada). Volume rendering and multiplanar reconstructions were performed using Amira 5.3 software (Mercury Computer System Chelmsford, MA, USA) (Pedullà et al., 2016). Root canal volume was calculated according to segments based on gray-level differences and determined using the software surface area function. Root canal paths were evaluated with high-resolution 3D rendering and orthogonal cross sections to verify root canal surface area and volume homogeneity of the groups at baseline.

### 2.2 | Root canal preparation

Teeth were randomly assigned to the Procodile or R6 Reziflow instrumentation technique. Specimens were decoronated to a standardized length of 13 mm (Pedullà et al., 2016). Additionally, small grooves were incised on the root surface with a round diamond bur to achieve a better superimposition within  $\mu$ CT images (Stern et al., 2012). The working length (WL) was determined as 0.5 mm shorter than the extension of a size 10 stainless steel hand file inserted into the root canal up to the apical foramen (Pedullà et al., 2016; Saber et al., 2017). Each root canal was initially shaped with the size 15, 0.3 taper Path glider (Komet) at 300 rpm. Then Procodile (size 25, .06 taper) or R6 Reziflow (size 25, .06 taper) were used in reciprocating motion (Reciproc ALL program using Wave one endo motor, Dentsply Maillefer, Baillagues, Switzerland) to the established WL. Each instrument was used for shaping two root canals. The root canals were irrigated between instruments with 0.5 mL of 1% sodium hypochlorite and 0.5 mL of 17% ethylenediaminetetraacetate alternate. Following instrumentation, a recapitulation with a size 10 hand stainless steel file and final irrigation with water were performed.

## 2.3 | Post-instrumentation micro-CT analysis

All specimens were scanned following instrumentalisation, as described previously. To enable calculation of the root canal volume, the same rendering settings were applied, and the canal volume was determined using the surface area function. Root canals were virtually divided into apical, middle, and coronal thirds, determined by the number of cross-sectional slices from the apex of the tooth to the full 12.00 mm reference point divided by 3 (Pedullà et al., 2016; Pedullà et al., 2019; Stern et al., 2012).

## 2.4 | Measurements of transportation and centring ratio

The 3D viewer and analysis imaging software MicroView was used to produce two-dimensional cross-sectional images of the samples. The centring ability (C) and canal transportation (T) were calculated using the following formula:

- if  $(D1 - D2) > (M1 - M2)$ :  $C = (M1 - M2)/(D1 - D2)$
- if  $(M1 - M2) > (D1 - D2)$ :  $C = (D1 - D2)/(M1 - M2)$
- $T = (M1 - M2) - (D1 - D2)$  (Gambill et al., 1996)

The shortest distances were considered between the boundary of the unprepared root canal to the mesial root aspect (M1), the periphery of the shaped canal to the mesial root (M2), the unprepared canal to the distal aspect of the root (D1), the distal part of the root to the periphery of the prepared root canal (D2) (Saber et al., 2017). These distances were calculated for each mesial root canal in all cross-sectional images from the floor of the pulp chamber to the apex (~350 slices for each root canal).

## 2.5 | Measurements of volume, surface area and unprepared area

Three-dimensional models of the root canals, before preparation and after shaping with Procodile and R6 Reziflow files, were matched and micro-CT scans were managed as previously described. Morphological parameters of the mandibular molar canals (volume, surface area, unprepared area) were acquired importing the TIFF converted cross-sectional images from the MicroView software into the 3D visualization and analysis software Amira 5.3 (Mercury Computer System Chelmsford, MA, USA). Measurement of canal and surface area volumes of the canals before and after instrumentation was evaluated separately, to precisely match the areas of interest through the AMIRA 5.3 software and its segmentation editor tool. In addition, the histogram tool was employed to calculate an automatic threshold, which was then used to produce an iso-surface (3-D  $\mu$ CT image) of the canal. Mean and SD of the volume and surface area increase ( $\Delta$ ) were expressed in  $\text{mm}^3$  and  $\text{mm}^2$ , respectively, for each root canal by subtracting the non-instrumented value from the instrumented one.

The mean percentage increase (% $\Delta$ ) and their relative SDs were also calculated. Spatially registered surface root models of the unprepared area of the root canal were also compared, calculated according to the distances between the surface of the root canals before and after preparation, determined at every surface point (Markvart et al., 2012). The percentage of the remaining unprepared surface area was calculated using the formula:

$$(Au/Ab) \times 100$$

Au represents the unprepared canal area and Ab the root canal area before preparation (Markvart et al., 2012). An examiner blinded to the preparation protocols performed all analyses.

## 2.6 | Statistical analysis

Results were presented as mean  $\pm$  SD for each group and evaluated with the Prism software (Prism 7.0; GraphPad Software, Inc, La Jolla, CA, USA). The Shapiro–Wilk test assessed data normality with significance level established at 5%. As data were normally distributed, the paired sample *t*-test was used to compare pre- and post-preparation parameters within groups. Statistical differences between groups were examined using the unpaired Student's *t*-test. Levene's Test for Equality of Variances was performed prior to the one-way analyses of variance (ANOVA) and the Tukey–Kramer post-hoc test was used to compare the mean values of different root canal thirds among groups.

# 3 | RESULTS

## 3.1 | Canal transportation

Significantly less transportation was observed with the Procodile compared to the R6 Reziflow instrumentation technique in the coronal third ( $p < .05$ ), while no differences were observed in the middle and apical thirds of the root, see Table 1.

## 3.2 | Centring ratio

There were no significant differences in any of the root canal thirds according to centring ability of instrumentation technique utilized, see Table 1.

## 3.3 | Volume, surface area and unprepared area

Pre-instrumentation geometrical parameters were revealed to be homogeneous between the two instrumentation technique groups. Overall, instrumentation significantly increased volume and surface area in both group ( $p < .05$ ). Qualitative assessment, represented by the superimpositions of unprepared (green) and prepared (red) areas,

**TABLE 1** Mean  $\pm$  SD of the canal transportation and centring ability after instrumentation with Procodile and R6 Reziflow instruments.

Experimental group	Canal transportation			Centring ability		
	Coronal third	Middle third	Apical third	Coronal third	Middle third	Apical third
Procodile	0.12 $\pm$ 0.39 <sup>a</sup>	0.07 $\pm$ 0.21	0.12 $\pm$ 0.11	0.26 $\pm$ 0.40	0.57 $\pm$ 0.28	0.49 $\pm$ 0.20
Reziflow	-0.56 $\pm$ 0.73	-0.04 $\pm$ 0.17	0.03 $\pm$ 0.14	0.41 $\pm$ 0.19	0.62 $\pm$ 0.40	0.66 $\pm$ 0.33

Note: Negative values indicate a transportation toward the distal aspect of the root.

<sup>a</sup>Statistically significant.

showed that the original canal shape remained unchanged, see Figure 1. The mean percentage increase of surface area was significantly higher with the R6 Reziflow instrument compared to the Procodile instrument in the coronal and middle thirds ( $p < .05$ ). The mean percentage volume increase and remaining unprepared surface area were not significantly different according to instrumentation techniques, see Table 2. The different radicular thirds were similarly not significantly different according to the instrumentation techniques utilized, see Table 3.

## 4 | DISCUSSION

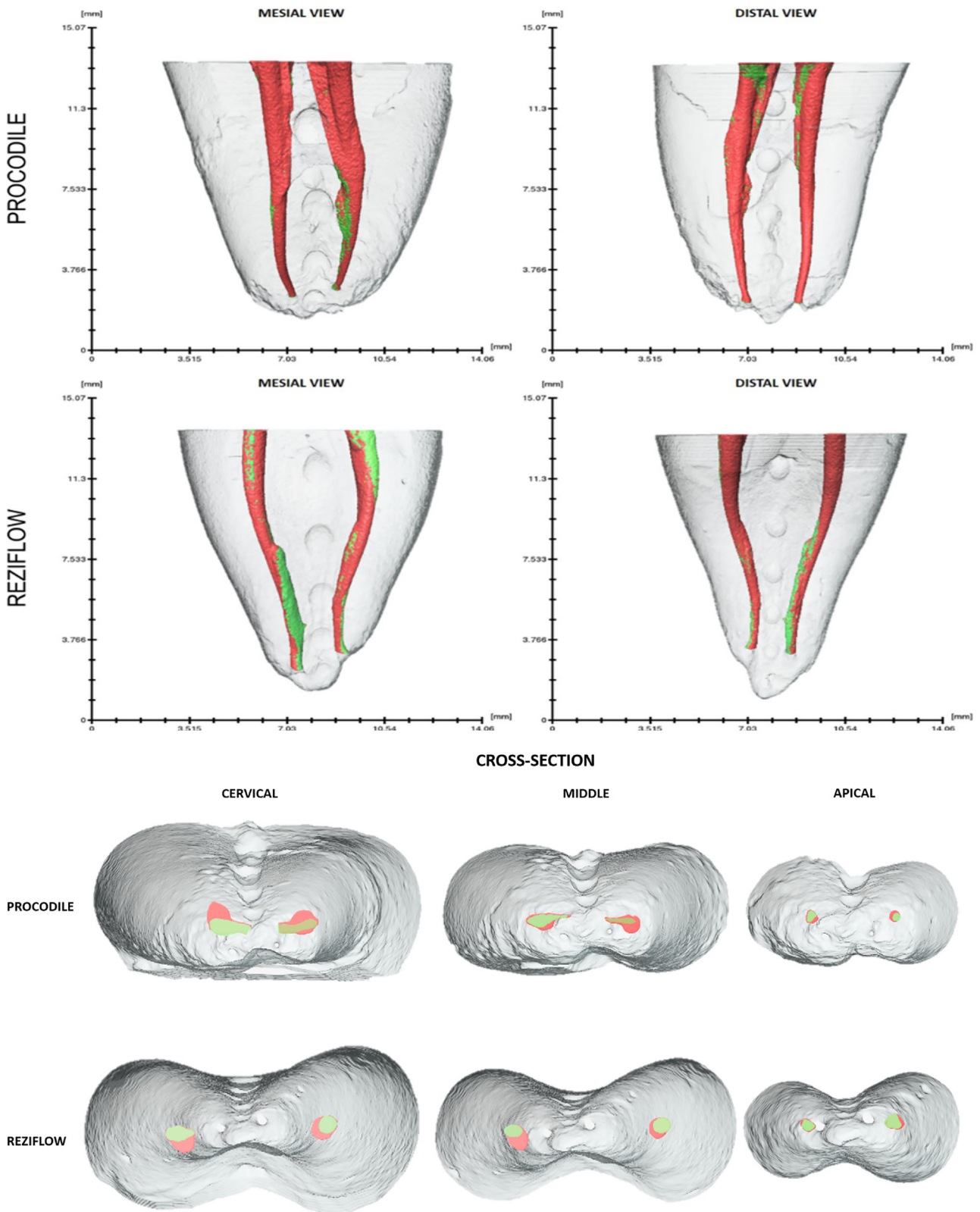
The Procodile and R6 Reziflow instruments, activated by the same reciprocating motor in this study, revealed a similar shaping ability when applied to root canals, with the only geometric differences observed with  $\mu$ CT evident in less coronal canal transportation and lower percentage increase of surface area with the Procodile compared to the R6 Reziflow.

Both Procodile and R6 Reziflow were recently introduced to the market and their shaping abilities and impact on the root canal morphology have not been extensively studied or compared with other instruments. Our investigation methodology was effectively designed to enable a reliable direct comparison between these instruments. As both instruments have a similar tip size, a reliable comparison of root canal morphology pre- and post-instrumentation was possible. Further, our study protocol specified the activated of both instruments with the same reciprocating motor, thereby eliminating any potential bias of different movements. Moreover, no orifice opener was employed to standardize methodological conditions. The  $\mu$ CT imaging ensured the standardization of samples in terms of preoperative geometric parameters (Alovisi et al., 2020; Gagliardi et al., 2015; Versiani et al., 2018), minimizing any potential anatomic biases (Versiani, Leoni, et al., 2013), while producing high-quality images. We were able to analyze specimens from the outer to the innermost structure and to evaluate the effect of different instrumentation on root canal preparation. With a special mounting device, we ensured an almost exact reposition of pre- and post-instrumentation images. Accuracy was also increased by varying the relative translation in different spatial axes rather than one voxel. Superimposition was both detected manually and with a dedicated software for images analyses (Neves et al., 2014). The great advantage of this investigation technique is that the specimen remains intact, and the same image can be viewed multiple times (Alovisi et al., 2022). Moreover, the specimen remains

available for further scanning for biological and mechanical testing, as well as accurate imaging reproduction (Neves et al., 2014).

When analyzed with the  $\mu$ CT, both instruments produced some degree of canal transportation but R6 Reziflow instruments produced a more relevant transportation in the coronal part of the root canal, which may be due to its greater cross-sectional area compared to Procodile, especially close to the shaft (Generali et al., 2020). Nevertheless, none of the instrumentation systems caused an apical transportation of more than 0.3 mm, which represents the experimental limit beyond which the seal of the obturation material might be compromised (Wu, Fan, & Wesselink, 2000; Wu, R'oris, et al., 2000). The centring ability of the instruments was not different between the root canal thirds, probably due to the similar stiffness of the two instruments (Generali et al., 2020). Further, no difference in the percentage increase of the volume was detected, while the percentage increase of surface area associated with the R6 Reziflow system exhibited significantly higher values than Procodile in coronal and middle regions. These results may be explained by the similar dimensions of the instruments (the same S cross-section, the same taper and tip diameter) but the different core area (Generali et al., 2020). The R6 Reziflow instruments have a greater core area than Procodile (Generali et al., 2020) which could cause less flexibility and consequently larger contact between the R6 Reziflow instrument and the root canal walls. According to the present results, Procodile and R6 Reziflow systems were associated with a relatively high mean percentage of unprepared canal walls, with no significant differences between the canal thirds. Currently, studies reporting the shaping ability of Procodile and R6 Reziflow are not yet available and thus a comparison with other results is not possible. However, all NiTi files have been associated with a variable degree of untouched area (Gagliardi et al., 2015). The flexibility of Procodile files, due to their reduced core area, seems not to enhance the shaping ability of the instruments probably caused by the small tip diameter and taper of the chosen files (size 25, .06 taper). Larger taper and bigger tip diameter should more influence the Procodile shaping ability exhibiting differences between these files and the equivalent R6 Reziflow instruments.

In this study, the mesial root canals of mandibular molars were prepared up to 25 apical diameter as in several previous study (Gagliardi et al., 2015; Lima et al., 2020; Pinheiro et al., 2017; Silva et al., 2022). The mean physiological foramina diameters of mesial root canals of first mandibular molars range from 0.24 to 0.41 mm (Wolf et al., 2017), and especially the mesio-buccal canals often present an oval anatomy (Wu, Fan, & Wesselink, 2000; Wu, R'oris, et al., 2000). Perez et al. (2018) demonstrated that as the instrument



**FIGURE 1** Mesial and distal 3D reconstructions and cross-sectional views of the cervical, middle and apical thirds of two representative mandibular molars before (in green) and after (in red).

size increases, the percentage of unprepared areas decreases significantly. Therefore, the main reason for which areas of the canal remain untouched is that the last shaping instrument is narrower than the

diameter of the canal in its largest part (Almeida et al., 2019). It is necessary to take into account that canals over-enlargement should be avoided because there is an increased risk of excessive removal of



**TABLE 2** 3D morphometric data of root canal system (mean  $\pm$  SD) before and after preparation using Procodile and ReziFlow.

Parameters	Instrument	
	Procodile	ReziFlow
Volume (mm <sup>3</sup> )		
Before	1.42 $\pm$ 0.32 <sup>a</sup>	1.74 $\pm$ 0.48 <sup>a</sup>
After	2.79 $\pm$ 0.54 <sup>a</sup>	4.07 $\pm$ 1.40 <sup>a</sup>
Increase ( $\Delta$ )	1.37 $\pm$ 0.46 <sup>a</sup>	2.33 $\pm$ 1.59 <sup>a</sup>
Increase ( $\Delta\%$ )	100.77 $\pm$ 45.12 <sup>a</sup>	157.29 $\pm$ 140.27 <sup>a</sup>
Surface area (mm <sup>2</sup> )		
Before	19.74 $\pm$ 3.90 <sup>a</sup>	19.41 $\pm$ 3.13 <sup>a</sup>
After	24.38 $\pm$ 4.06 <sup>a</sup>	30.32 $\pm$ 6.16 <sup>b</sup>
Increase ( $\Delta$ )	4.63 $\pm$ 1.87 <sup>a</sup>	10.90 $\pm$ 6.81 <sup>b</sup>
Increase ( $\Delta\%$ )	24.65 $\pm$ 12.62 <sup>a</sup>	59.27 $\pm$ 40.84 <sup>b</sup>
Unprepared area (mm <sup>2</sup> )		
After	5.59 $\pm$ 3.06 <sup>a</sup>	6.54 $\pm$ 3.15 <sup>a</sup>
After (%)	29.10 $\pm$ 16.62 <sup>a</sup>	34.27 $\pm$ 18.17 <sup>a</sup>

Note: Different superscript letters in the same line indicate a statistically significant difference between groups ( $p < .05$ ).

tooth structure which could lead to root fracture (Almeida et al., 2019). Current rotating or reciprocating instruments dimensions are not able to completely prepare the mesial canals of mandibular molars. The ideal instrument size/taper able to engage all canal walls would be too large (Almeida et al., 2019) and there is no unanimous consensus in the literature regarding the ideal diameter of apical preparations (Pinheiro et al., 2017). In this study, it was decided not to use instruments with a larger tip diameter and the same taper to avoid excessively weakening the tooth structure with narrow canals. An apical enlargement greater than #25 in the mesial canals of mandibular molars is preferable for the removal of bacteria from the apical area. Instruments with less than 6% taper can avoid excessive weakening of the tooth structure. Finally, on the basis of our findings, the null hypothesis can be partially rejected. The present results are limited to the type of instrument and anatomical configuration investigated and should not be generalized. Indeed, shaping outcomes derive from different variables including file geometry, kinematics, operator experience, and motor setting employed during root canal shaping (Zuolo et al., 2018). The current results are clinically relevant because they show that, despite both files allowing an acceptable root canal preparation, no instrument can ensure an ideal preparation of a root canal system. Irrigation is thus pivotal to compensate for the suboptimal status of mechanical preparation (Versiani, Leoni, et al., 2013).

## 5 | CONCLUSIONS

Procodile and R6 ReziFlow systems showed a similar shaping ability. Procodile had lower root canal transportation values in the coronal third. Regarding the percentage increase of surface area, R6 ReziFlow

**TABLE 3** 3D morphometric data (mean  $\pm$  SD) before and after instrumentation with Procodile and ReziFlow instruments referred to each root canal third.

Experimental group	Root third	Volume (mm <sup>3</sup> )			Surface area (mm <sup>2</sup> )			Unprepared area (mm <sup>2</sup> )		
		Before	After	Increase ( $\Delta$ )	Before	After	Increase ( $\Delta$ )	Before	After	Increase ( $\Delta$ )
Procodile	Coronal	0.76 $\pm$ 0.21	1.49 $\pm$ 0.25	0.73 $\pm$ 0.27 <sup>a1</sup>	9.57 $\pm$ 2.19	11.64 $\pm$ 2.58	2.07 $\pm$ 1.02 <sup>a1</sup>	3.19 $\pm$ 1.83 <sup>a1</sup>	36.38 $\pm$ 23.98	3.19 $\pm$ 1.83 <sup>a1</sup>
	Middle	0.46 $\pm$ 0.13	0.89 $\pm$ 0.19	0.42 $\pm$ 0.14 <sup>a1</sup>	6.76 $\pm$ 1.23	8.09 $\pm$ 1.29	1.33 $\pm$ 0.56 <sup>a1</sup>	1.59 $\pm$ 1.05 <sup>a1</sup>	25.39 $\pm$ 18.76	1.59 $\pm$ 1.05 <sup>a1</sup>
	Apical	0.19 $\pm$ 0.06	0.40 $\pm$ 0.12	0.20 $\pm$ 0.10 <sup>a1</sup>	4.11 $\pm$ 1.05	5.40 $\pm$ 1.03	1.29 $\pm$ 0.75 <sup>a1</sup>	1.25 $\pm$ 0.76 <sup>a1</sup>	28.83 $\pm$ 14.67	1.25 $\pm$ 0.76 <sup>a1</sup>
ReziFlow	Coronal	1.01 $\pm$ 0.28	2.38 $\pm$ 0.76	1.36 $\pm$ 0.86 <sup>b1</sup>	9.49 $\pm$ 2.03	15.67 $\pm$ 2.29	6.17 $\pm$ 3.01 <sup>b2</sup>	2.91 $\pm$ 2.08 <sup>b1</sup>	31.24 $\pm$ 25.35	2.91 $\pm$ 2.08 <sup>b1</sup>
	Middle	0.47 $\pm$ 0.21	1.22 $\pm$ 0.50	0.75 $\pm$ 0.53 <sup>b1</sup>	6.05 $\pm$ 1.86	9.57 $\pm$ 2.84	3.52 $\pm$ 2.79 <sup>b2</sup>	1.46 $\pm$ 1.05 <sup>b1</sup>	23.23 $\pm$ 14.17	1.46 $\pm$ 1.05 <sup>b1</sup>
	Apical	0.25 $\pm$ 0.12	0.50 $\pm$ 0.14	0.25 $\pm$ 0.21 <sup>b1</sup>	4.28 $\pm$ 0.65	5.74 $\pm$ 1.67	1.45 $\pm$ 1.57 <sup>b1</sup>	1.88 $\pm$ 1.15 <sup>b1</sup>	41.64 $\pm$ 18.59	1.88 $\pm$ 1.15 <sup>b1</sup>

Note: Different superscript letters in the same column indicate a statistically significant difference among the root canal thirds for the same instruments ( $p < .05$ ). Different superscript numbers in the same column indicate a statistically significant difference among the two instruments tested ( $p < .05$ ).

showed better results with an increase of the canal surface touched by the files. Both Procodile and R6 Reziflow do not seem to substantially modify the original tooth anatomy and could be safely used in curved root canals.

## AUTHOR CONTRIBUTIONS

**Luigi Generali:** Conceptualization; funding acquisition; methodology; project administration; supervision; writing – original draft. **Vittorio Checchi:** Formal analysis; writing – review and editing. **Alessia Borghi:** Formal analysis; investigation; methodology; writing – original draft. **Giusy Rita Maria La Rosa:** Writing – original draft; writing – review and editing. **Gianluca Conte:** Formal analysis; investigation; methodology. **Angelo Zavattini:** Formal analysis; investigation; software; visualization; writing – original draft. **Francesco Mannocci:** Conceptualization; supervision; validation; writing – review and editing. **Daniele Angerame:** Methodology; resources; software. **Ugo Consolo:** Supervision; validation; writing – review and editing. **Eugenio Pedullà:** Conceptualization; validation; writing – original draft.

## ACKNOWLEDGMENTS

The authors report that the research was funded from the FAR project (University of Modena and Reggio Emilia Funds for Research) entitled Microstructural and Mechanical investigation of Nickel-Titanium Endodontic instruments—MMiNTEndo. The authors are grateful to Dr. Christopher Healy for the execution of the  $\mu$ CT, Prof. Francesco Cavani for the support in the statistical analysis and Johanna Chester for editing the manuscript. Open Access Funding provided by Università degli Studi di Modena e Reggio Emilia within the CRUI-CARE Agreement.

## CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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**How to cite this article:** Generali, L., Checchi, V., Borghi, A., La Rosa, G. R. M., Conte, G., Zavattini, A., Mannocci, F., Angerame, D., Consolo, U., & Pedullà, E. (2023). Shaping ability of Procodile and R6 Rezipflow nickel-titanium reciprocating instruments in curved mesial root canals of mandibular molars: A MicroCT study. *Microscopy Research and Technique*, 86(10), 1345–1352. <https://doi.org/10.1002/jemt.24326>