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Micro-computed tomographic evaluation of the effect of the final apical size prepared by rotary nickel-titanium files on the removal efficacy of hard-tissue debris

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Abstract

Objective: This study was performed to compare the removal efficacy of hard-tissue debris in mandibular first molars prepared to various apical sizes.

Methods: The mesial root canals of 40 extracted mandibular first molars were prepared by rotary nickel-titanium files to an apical size of #25/0.04 (Group #2504), #30/0.04 (Group #3004), #35/0.04 (Group #3504), and #40/0.04 (Group #4004). Irrigation was performed with 5.25% NaOCl and 17% EDTA. Micro-computed tomography was used to scan the samples before and after root canal preparation. Remnant hard-tissue debris was identified, quantified, and visualized three-dimensionally using shape recognition and image superimposition analysis, and the debris was mapped to its location.

Results: After preparation, 11.67%±2.99% of the root canals contained hard-tissue debris in Group #2504, 8.00%±2.71% in Group #3004, 7.17%±2.88% in Group #3504, and 4.51%±1.61% in Group #4004. The amount of accumulated hard-tissue debris was significantly less in Group #4004 than in the other groups. However, there were no significant differences between Groups #3004 and #3504.

Conclusions: An increased final apical size resulted in significantly lower debris accumulation. However, no root canal in any group was completely free from hard-tissue debris, and debris was mostly found in the isthmus of the mesial root canals.

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Introduction

Hard-tissue debris produced by mechanical root canal shaping with rotary nickel-titanium (NiTi) files can be partially removed by simultaneous chemical irrigation.[1] Because of the complexity of the root canal system, small amounts of mixed debris composed of dentin particles, necrotic pulp tissue, and pathogenic microorganisms are resistant to clearance.[2,3] Brito *et al.*[4] reported that residual debris after chemical and mechanical preparation can accumulate in irregular areas of the root canal system such as the apical delta and isthmus. Thus, the remaining debris poses a greater problem than the smear layer, which can facilitate the formation of a bacterial biofilm and deteriorate the sealing ability of root canal filling materials. Additionally, residual bacteria may proliferate and reinfect the root canal system, hindering the healing of endodontic lesions or resulting in root canal treatment failure.[5–7]

Debris cannot be quantitatively analyzed by traditional methods, such as scanning electron microscopy, traditional radiography, or cone-beam computed tomography (CBCT). Scanning electron microscopy is commonly performed on two-dimensional cross sections of teeth, which provides a limited view of where the debris is accumulated.[8–10] CBCT is clinically applied because of its capacity to reveal the three-dimensional (3D) morphology of root canals. However, the resolution of CBCT restricts its use in analyzing the complex root canal morphology.[11] Paque *et al.*[12] proposed a new method for *in vitro* quantitative analysis of debris using micro-computed tomography (micro-CT) scans before and after root canal preparation. Micro-CT can create 9-µm cross-sectional images and is widely used in laboratory research of bones and teeth. Micro-CT techniques enable 3D reconstruction of the root canal system. Additionally, its non-destructibility and reproducibility make it possible to compare root canal parameters before and after root canal preparation, such as the root canal volume, surface area, and deviation.[11] Moreover, micro-CT techniques have been used in some studies to analyze the debris removal efficacy of NiTi instruments with specific designs.[6,13–15]

Previous studies have shown that the increased size and taper of the master apical file improve debris and smear layer removal.[16–18] Furthermore, Silva et al.[19] evaluated the amount of apical debris extruded by four systems and found that all systems were associated with apical debris extrusion when canals were prepared to a large apical size. Because the anatomy of the root canal system varies by ethnic populations, it is essential to investigate the correlation between the final apical preparation size and the efficacy of debris removal in Chinese populations. The iRace NiTi instrument (FKG Dentaire, La-Chaux-de-Fonds, Switzerland) is the simplified sequence of the Race system (FKG Dentaire) and has been newly promoted in the Chinese market. The iRace system, which contains an electrochemical polishing cutting edge, reportedly provides efficient cutting and torque resistance. The triangular cross section increases the flexibility of the system and allows for easier discharge of debris.[20] However, the effectiveness of the files has not been evaluated in mandibular molar teeth of Chinese individuals.

The aim of this study was to explore the relationship between the amount of debris remaining after root canal preparation by the iRace and the apical preparation size in mesial root canals of mandibular first molars.

Materials and methods

Sample collection

Mandibular first molars were collected from patients of the Oral Surgery Clinic (Department of Oral Surgery, Affiliated Hospital of Stomatology, Nanjing Medical University) after approval by the Ethics Committee of Nanjing Medical University (PJ2014-029-001). Information about the patients' Chinese nationality was confirmed. All patients provided written informed consent. All selected teeth were extracted over a 3-month period and had a complete tooth structure and fully formed apices. No included teeth had apical absorption or cracks on the tooth surface or had received endodontic treatment under microscopy (Leica 400 microscope; Leica, Wetzlar, Germany). All teeth were stored in individual glass jars containing 0.1% thymol solution before experimentation. After all attached soft tissues and calculi were removed with a sterile scalpel, all selected teeth were scanned by CBCT. The curvature angle was measured using an image analysis program (Mimics v.10.0; Materialise NV, Leuven, Belgium), and only teeth with a mesial root with moderate curvature (ranging from 10°–20°) were selected. Only mandibular first molars with two independent mesial root canals (Vertucci type II configuration system[21]) were included for analysis.

Micro-CT scanning and grouping

The 40 mandibular first molars were placed crown-down inside a custom-made silicone rubber holder that could keep all samples in the same position; they were then scanned using a micro-CT device (SkyScan 1176; Bruker microCT, Kontich, Belgium). Scanning procedures were performed at a resolution of 9 μm, 90 kV, 278 mA, 360° rotation around the vertical axis, and rotation step of 0.24°. The cross-sectional image was reconstructed from the projection image with a beam-hardening compensation of 45%. Cross-sectional images were segmented, registered, visualized, and quantified using CTAn v.1.14 and CTVol v.2.2.3 software (Bruker microCT). The maximum root canal diameter (D) 1 mm from the root apices was measured and recorded. According to obtained D values, the 40 mandibular first molars were randomly assigned to 4 groups ($n = 10$ each). The D values were not significantly different among the four groups.

Root canal preparation

A stainless steel size 10 hand k-file (Dentsply Maillefer, Ballaigues, Switzerland) was inserted into the canal up to the apical foramen. The working length was confirmed at 1 mm from the length of the canal. A single experienced operator performed two mesial root canal preparations with iRace files (FKG Dentaire) according to the manufacturer's instructions. Apical enlargements were prepared to apical sizes of #25/0.04 (Group #2504), #30/0.04 (Group #3004), #35/0.04 (Group #3504), and #40/0.04 (Group #4004) using the following sequence of instruments: 15/0.06 working length (WL), 25/0.04 WL, 30/0.04 WL, 35/0.04 WL, and

40/0.04 WL. The X-Smart (Dentsply Maillefer) electric motor was used with the rotation speed adjusted to 250 rpm and 1 Ncm torque. After four gentle in-and-out strokes, the instrument was removed from the canal, and the canal was cleaned until the WL was reached.

According to previously published work,[2,22] the root canals were irrigated with 2 mL of 5.25% NaOCl for 1 minute, delivered by a side-vented irrigating tip (Max-i-Probe; Hawe-Neos, Dentsply, Bioggio, Switzerland) placed 2 mm short of the WL of each rotary instrument. At the end of the preparation, the canals were flushed with 10 mL of 5.25% NaOCl for 5 minutes, followed by 2 mL of 17% EDTA for 5 minutes. The canals were then dried with paper points.

Micro-CT scanning after root canal preparation

After root canal preparation, the teeth were scanned by micro-CT using the same silicone rubber holder and selected parameters as before root canal preparation. The acquired projection images were reconstructed into cross-sectional slices (NRecon v.1.6.9; Bruker microCT) using standardized parameters for beam hardening (40%) and a ring artifact correction of 10 with similar contrast limits. The volume of interest extended from the furcation level to the apex of the mesial root, resulting in the acquisition of 700 to 900 transverse cross sections per tooth. The data were then analyzed by CTAn and CTVol software (Bruker microCT).

Quantitative 3D image analysis

The evaluation procedures used in the present study have been described in detail elsewhere.[3] Image stacks were registered before and after root canal preparation using automatic superimposition, and the volumes of the matched root canals before and after preparation were calculated. Materials with a density similar to dentin in the instrumented canal regions previously occupied by air were considered to be debris.[23] The amount of accumulated hard-tissue debris was calculated by the percentage volume of the original canal anatomy after intersecting the stacks of the original volume and the instrumented root canal space. All image analysis operations were undertaken using CTAn software. The images obtained after debris quantification were then qualitatively evaluated using CTVol v.2.2.1 software (Bruker microCT).

The result of the quantification process was a set of images containing the root canal space and images containing debris. The canal space and debris volumes were determined through voxel summation.

The percentage of debris remaining was calculated as follows: $t = d3/(d1 + d2) \times 100$, where t is the percent of total debris accumulated after instrumentation and irrigation, d1 is the debris before preparation, d2 is the debris created during preparation, and d3 is the debris present in the preparation. Debris volumes before and after preparation were acquired from processed data. The debris created during preparation was proportional to and acquired from changes in the canal space volume. CTAn software was used to generate models that were analyzed using CTVol software.

Statistical analysis

Measurement data are expressed as the mean ± standard deviation. Normal distribution of the raw data was confirmed using the Shapiro–Wilk test ($p > 0.05$). The percentage volume of accumulated hard-tissue debris after preparation was compared using one-way analysis of variance with the Student–Newman–Keuls test for between-group variance. A two-sided

p value of <0.05 was considered statistically significant for all analyses. IBM SPSS Statistics for Windows, Version 19.0 (IBM Corp., Armonk, NY, USA) was used for all statistical analyses.

Table 1. Volume (mm^3) of dentin removed and percentage volume of accumulated hard tissue after preparation of mesial canals of Chinese mandibular molars with different apical enlargement sizes.

Groups	Volume (mm^3) of dentin removed	Volume (%) of accumulated hard tissue
#2504	1.61 ± 0.36	11.67 ± 2.99
#3004	2.11 ± 0.66	8.00 ± 2.71
#3504	2.58 ± 0.89	7.17 ± 2.88
#4004	2.83 ± 0.77	4.51 ± 1.61

Data are presented as mean ± standard deviation.

Results

In total, 40 mandibular first molars with 2 independent mesial root canals were analyzed. Table 1 shows the volume of dentin cut by root canal preparation and the percentage volume of accumulated hard-tissue debris in the mesial canals of all Chinese mandibular first molars with various apical enlargement sizes. Figure 1 shows the percentage volume of accumulated hard-tissue debris in the four groups: 11.67%±2.99% (Group #2504), 8.00% ±2.71% (Group #3004), 7.17%±2.88% (Group #3504), and 4.51%±1.61% (Group #4004). The percentage of accumulated hard-tissue debris during mesial canal preparation was significantly different among the groups ($p < 0.05$). The amount

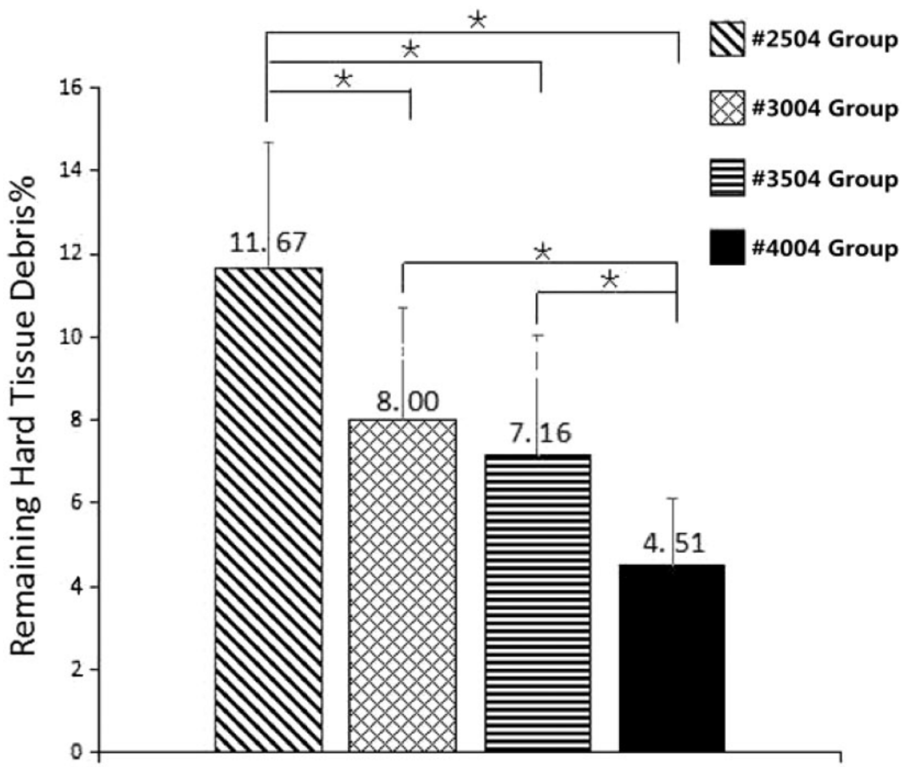

Figure 1. Mean percentage volume and standard deviation of hard-tissue debris in each experimental group. *$p < 0.05$.

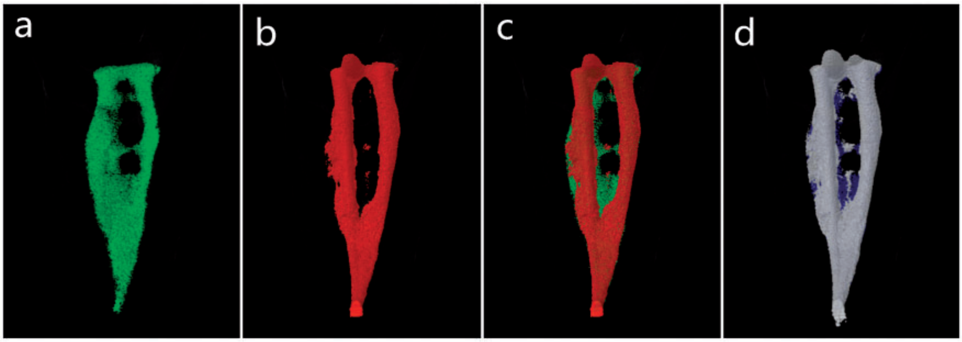

Figure 2. Three-dimensional reconstruction of micro-computed tomography (micro-CT) scans of the mesial root canal system of Chinese mandibular first molars investigated in this study. (a) The initial canal configuration with the complex isthmus area from its coronal part to apical part. (b) Three-dimensional micro-CT reconstruction after instrumentation. (c) Corresponding three-dimensional micro-CT reconstruction after instrumentation. Prepared canal areas are indicated in red, and untouched areas are indicated in green. (d) Superimposition of apparent accumulated hard-tissue debris areas is indicated in blue. The canal space and empty space in the isthmus after instrumentation are indicated in gray.

of accumulated hard-tissue debris in Group #4004 was significantly lower than that in the other three groups ($p < 0.05$), and the amount of debris in Group #2504 was significantly greater than that in the other three groups. However, there were no significant differences between Groups #3004 and #3504. Figure 2 shows representative 3D reconstructions of the mesial root canals of Chinese mandibular first molars before and after preparation; the accumulated hard-tissue debris with three different systems visually agreed with the quantitative results. Figure 3 shows cross-sectional images of accumulated hard-tissue debris in the mesial root canals of Chinese mandibular first molars. In total, 29 (72.5%) mandibular first molars with isthmuses in the mesial canals were assessed in this study. More debris remained in these molars, particularly in the isthmuses.

Discussion

A smear layer and/or hard-tissue debris is inevitably created on the surface of root canal walls when NiTi instruments are used for mechanical preparation. The smear layer, which is loosely accumulated, can be easily removed using an irrigant such as EDTA.[24,25] Most studies have suggested that bacteria inside the smear layer can be eliminated and are therefore not critical to the outcome of root canal treatment.[26,27] However, hard-tissue debris created after root canal preparation has been regarded as more relevant to the success of root canal therapy than the smear layer because it can harbor bacterial contaminants even after disinfection.[12] Bacteria and their metabolites can gain access to periapical tissues through the apex and dentinal tubules. Bacterial invasion of dentinal tubules can reportedly reach 300 to 1000 μm, whereas the penetration of infected root canals by an irrigant such as NaOCl is limited to approximately 130 μm. Thus, it is impossible to eliminate deep infection that is far from the inner root canal wall.[28] Many studies have proposed that additional enlargement of the terminal preparation size of the apical portion of the root canal could improve the root canal cleaning efficiency and maximize disinfection of the root canal system.[29–31] Aminoshariae and Kulild[32] reviewed relevant literature from

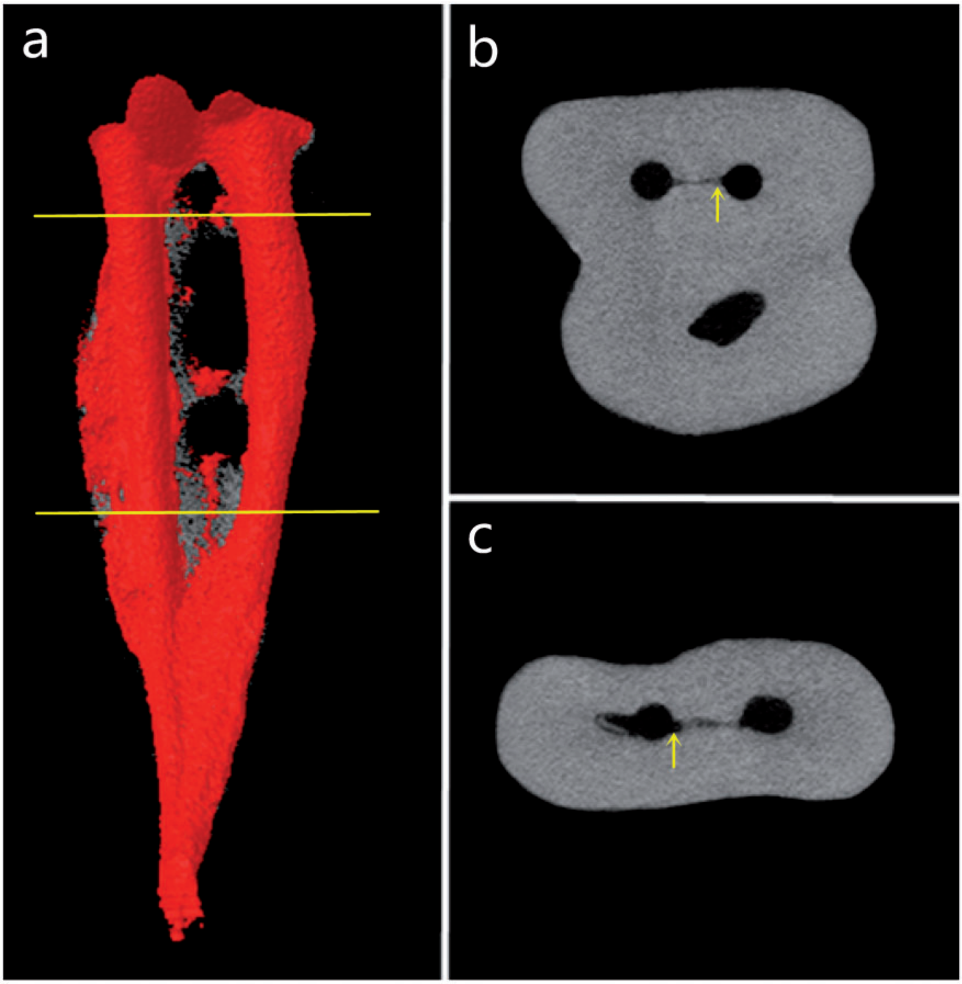

Figure 3. Three-dimensional and cross-sectional image reconstruction of micro-computed tomography (micro-CT) scans of accumulated hard tissue after preparation in mesial canals of mandibular molars. (a) Three-dimensional micro-CT reconstruction of accumulated hard tissue. (b) Two-dimensional micro-CT reconstruction of accumulated hard tissue. The arrow refers to the first imaginary line (in yellow) in (a). (c) Two-dimensional micro-CT reconstruction of accumulated hard tissue. The arrow refers to the second imaginary line (in yellow) in (a).

the past 60 years and concluded that it is impossible to obtain a completely clean root canal with the existing chemical preparation methodologies, regardless of the width of root canal preparation. Thus, exploring the appropriate width of root canal preparation remains a clinical interest.

The use of micro-CT scans before and after root canal preparation *in vitro* along with professional software to analyze the level of residual debris was first described by Paque *et al.*[12] in 2009 and was well-defined by Robinson *et al.*[23] in 2012. De-Deus *et al.*[2] recently compared the amount of hard-tissue debris produced after apical enlargement with two single-file reciprocating systems (WaveOne; Dentsply Maillefer and Reciproc; VDW, Munich, Germany) and a conventional multi-file rotary system (BioRace; FKG Dentaire) using micro-CT scans. The authors found that none of the systems yielded root canals completely free from packed hard-tissue debris; however, an increased final apical size resulted in significantly less debris accumulation for both reciprocating and rotary systems. Paque *et al.*[33] used a similar method and found that irrigation with 2.5% NaOCl resulted in 5.5%$\pm$3.6 vol% accumulated hard-tissue debris compared with 3.8%$\pm$1.8 vol% when etidronic acid (HEDP) was contained in the irrigant ($p < 0.05$). In the present study, to determine the most suitable root preparation width of mandibular first molar mesial roots and provide a clinical reference for Chinese populations, we compared the removal of residual debris using the same NiTi files to prepare the root canal to different apical canal widths.

After preparing the mesial canals of 40 teeth to different apical sizes with iRace, we found that the more dentin was cut, the less residual debris remained in the canals. There was no significant difference between Groups #3004 and #3504; however, the amount of residual debris in Group #4004 was significantly reduced ($p < 0.05$). This finding explains why although more debris was created when the canal was prepared to a larger apical size, the volume of the root canal was increased, and the fluid was more likely to flow to the apical region. Thus, liquid reflux was increased, and the level of debris was reduced, leading to a cleaner canal. In fact, because of the irregular anatomy of the apical root region, it was difficult to thoroughly clean the root canals by simply increasing the root preparation size, and there was an increased risk of perforation and root fracture due to excessive cutting.[34] Therefore, none of the root canals in the four groups in our study were completely free of hard-tissue debris, and debris readily accumulated in the isthmus of the mesial root canals. Our results are consistent with those of Aminoshariae and Kulild[32], who reviewed 60 years of literature and found that a completely clean canal could not be obtained using the present chemical flushing methods. Brunson *et al.*[35] found that an increased width and taper of the apical preparation could lead to a significantly greater fluid volume. They also suggested that an apical region prepared to #40/0.04 was more conducive to irrigation of the apical third of the root canal. Similarly, de Gregorio *et al.*[36] found that the apical flushing fluid volume significantly increased when the size of the root tip preparation changed from #35/0.06 to #40/0.04.

According to published protocols used by Paque *et al.*[16] and Endal *et al.*,[5] irrigation with 5.25% NaOCl and 17% EDTA was used in these studies[37–39] to control the influence of chemical preparation on the experimental results with standard irrigation. However, even when the canals were prepared to a larger size (#40), we still found that debris remained after irrigation. This is similar to the findings of De-Deus *et al.*[40] Using micro-CT, they observed remnant debris in canals after preparing mandibular molars to different apical sizes with rotating or reciprocating files and found that neither system could thoroughly eliminate debris. When the apical size increased to #40, the remaining debris was decreased. Furthermore, De-Deus *et al.*[2] used three different approaches to irrigate the mesial root canals of mandibular molars and found that accumulated debris remained regardless of the irrigating protocol used, particularly in the isthmus. In the present study, we found an isthmus in 29 (72.5%) of the 40 molars, and these teeth had much more debris after root canal preparation than those without an isthmus. This result was consistent with the findings of Fan *et al*.[41] Similarly, other studies[42,43] have shown that debris might accumulate in the isthmus after root canal preparation. Because of the complex isthmus anatomy between the canals, it can be difficult to flush away debris once it has adhered to the canal wall of the isthmus.

Root canal preparation with large-taper NiTi files might reduce the resistance of roots and increase the risk of fracture.[44,45] Bier *et al*.[46] found that roots prepared with rotary files with a taper of $\geq$0.06 showed defects. They believed that the taper of files could contribute to the generation of dentinal defects, which could ultimately lead to tooth fracture. Race NiTi instruments use a taper of 0.04 as the working preparation taper to reduce over-cutting of dentin and increase fracture resistance. These files have good flexibility[47] and cyclic fatigue resistance[48] because of the physical design of a highly electrochemical polishing cutting edge and triangular cross section, leading to minimal canal transportation and maintenance of the original canal path. iRace is the simplified sequence of the Race system, which was recently introduced to the Chinese market. Saber *et al*.[49] recently compared the shaping ability of curved canals between the iRace and ProTaper Next. They found that the iRace resulted in less canal transportation and was safe to use.

Conclusion

In this study, we found that when the final apical size increased, less debris remained. However, none of the root canals of the four groups were completely free from hard-tissue debris, and debris readily accumulated in the isthmus of mesial root canals. When the apical size increased to #40, the remnant debris significantly decreased in the mesial roots of Chinese mandibular first molars.

Declaration of conflicting interests

The authors declare that there is no conflict of interest.

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