Clinical Oral Investigations (2019) 23:681–687
https://doi.org/10.1007/s00784-018-2483-1

ORIGINAL ARTICLE

Micro-CT evaluation of different final irrigation protocols on the removal of hard-tissue debris from isthmus-containing mesial root of mandibular molars

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Received: 7 January 2018 / Accepted: 2 May 2018 / Published online: 10 May 2018

Abstract

Objectives This study aimed to compare four final irrigation protocols (passive ultrasonic irrigation [PUI], EndoVac, Self-Adjusting File [SAF] and EasyClean) on the removal of accumulated hard-tissue debris (AHTD) from mesial canals of mandibular molars through microcomputed tomographic (micro-CT) analysis.

Materials and methods Forty mesial roots of mandibular molars presenting isthmuses type I or III were scanned in a micro-CT device and instrumented up to Reciproc R40 instrument. After the completion of canal preparations, root canals of each group were submitted to a final rinse using 20 mL of solution (16 mL of 5.25% NaOCl and 4 mL of 17% EDTA) in a total time of 5 min according to one of the four final irrigation protocols ($n = 10$): PUI, EndoVac, SAF and EasyClean operated at reciprocating motion. The sample was scanned again after canal preparation and after the use of the final irrigation protocols, and the registered data sets were examined to evaluate the percentage of AHTD. Data were statistically compared using the Tukey test with a significance level set at 5%.

Results All groups presented a decrease on the accumulation of hard-tissue debris after the use of the final irrigation protocols ($P < 0.05$). No significant differences in the removal of AHTD were observed among the final irrigation protocols ($P > 0.05$).

Conclusions All final irrigation protocols showed the same effectiveness in the removal of AHTD. None of them was able to render mesial canals of mandibular molars completely free from packed debris.

Clinical relevance This study highlighted that all final irrigation protocols (PUI, EndoVac, SAF, and EasyClean) promoted a similar removal of AHTD. However, none of the final irrigation protocols was able to render mesial canals of mandibular molars completely free from packed debris.

Keywords Debris · EasyClean · Micro-CT · Passive ultrasonic irrigation · Root canal irrigation

Introduction

During root canal preparation, the use of endodontic instruments on dentinal walls produces an accumulation of hard-tissue debris in anatomic irregularities of the root canal space [1–5]. This situation may be considered more clinically relevant in infected root canals than the smear layer per se, because its sizable amount can easily harbor microorganisms from the hydrodynamic action of irrigant flow [6]. In addition, it may also interfere with root canal filling materials from reaching difficult areas of the canal space [7].

As conventional irrigation performed by syringe and needle is generally inefficient on the removal of accumulated hard-tissue debris (AHTD) from anatomic irregularities, several supplementary approaches have been proposed to drive

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irrigants into such anatomic complexities of the root canal system [8–12]. The currently available techniques include the passive ultrasonic irrigation (PUI), which is characterized by the activation of an irrigant in the root canal using ultrasonically oscillating small files or specific tips. Its effectiveness on the removal of tissues and debris has been studied extensively [8]. Another supplementary approach is the EndoVac system (Discus Dental, Culver City, EUA), which generates negative pressure through a microcannula inserted within the vicinity of the working length (WL) and facilitates apical flow of the irrigant with minimal extrusion [9]. The Self-Adjusting File (SAF; ReDent-Nova, Ra'anana, Israel) is a hollow, cylinder-like, nickel-titanium motor-driven file that undertakes root canals preparation by means of scrapping canal walls with vertical vibrations. During this procedure, continuous irrigation throughout SAF instrument is also performed [10].

Recently, a new irrigation instrument (EasyClean; Easy Equipamentos Odontológicos, Belo Horizonte, MG, Brazil) was launched into the market. The EasyClean instrument presents a size of 25/0.04, an "aircraft wing"-shaped cross section and is made of an acrylonitrile butadiene styrene plastic designed to provide great flexibility, yielding vigorous intracanal fluid agitation [11, 12]. It has been demonstrated that this instrument is effective to promote penetration of irrigant solution into simulated lateral canals [11], providing a better removal of smear layer in the apical third when compared to PUI [12].

Despite these facts, comprehensive knowledge regarding the use of EasyClean in root canal debridement is still lacking. Therefore, this study compared four final irrigation protocols (PUI, EndoVac, SAF and EasyClean) on the removal of AHTD from mesial canals of mandibular molars through microcomputed tomographic (micro-CT) imaging. The null hypothesis tested was that there would be no differences in the removal of AHTD between the final irrigation protocols.

Materials and methods

Sample size estimation

According to a previous study with similar methodology [3], the effect size for this study was established (= 0.91) and added to a power $\beta = 95\%$ and $\alpha = 5\%$ inputs into an F test family for one-way analysis (G*Power 3.1.7 software for Windows; Heinrich Heine, Universität Düsseldorf). Twenty-six specimens were indicated as the ideal sample size required for observing significant differences. Additional specimens were used to compensate possible sample loss.

Selection and distribution of specimens

This study was approved by the local Ethics Committee (protocol 47,448,315.2.0000.5283). A total of 142 human first and second mandibular molars were selected and stored in a 0.1% thymol solution at 5 °C. Digital radiographs were then taken from each specimen in a buccolingual direction. This stage aimed to permit the estimation of the curvature angle of mesial roots according to Schneider's method [13] by the use of AxioVision 4.5 software (Carl Zeiss Vision GmbH, Hallbergmoos, Germany). Only teeth presenting moderately curved mesial roots (10–20°) were included, and, according to this criterion, 77 specimens were selected.

Selected specimens were scanned in a micro-CT device (SkyScan 1173; Bruker micro-CT, Kontich, Belgium) using the following parameters: 70 kV and 114 mA, pixel size of 14.25 μm, 360° rotation around the vertical axis, rotation step of 0.5°, frame averaging of 5 and a 1.0-mm-thick aluminum filter. The acquired projection images were reconstructed using NRecon v.1.6.10 software (Bruker micro-CT) to provide axial cross sections by using standardized parameters for beam hardening (30%), ring artifact correction of 5, and resembling contrast limits. In this study, the volume of interest covered the cementoenamel junction until the root apex, leading to the obtainment of 700–800 transverse cross sections per specimen. According to the three-dimensional models obtained, 40 teeth presenting mesial roots with isthmuses type I or III were selected. Isthmuses type I and III have a narrow sheet presenting a total union between two canals and an incomplete isthmus that is above or below a complete isthmus, respectively [14]. Afterwards, according to the final irrigation protocol, selected teeth were randomly assigned into one of the four experimental groups ($n = 10$): PUI, EndoVac, SAF, or EasyClean. Data normality ($P > 0.05$; Shapiro-Wilk) and homogeneity of groups were evaluated as regards to root length and degree of curvature, thus confirming anatomical matching between experimental groups ($P > 0.05$; two-way ANOVA).

Root canal preparation

After access cavity preparation, a size #10 K-file (Dentsply Maillefer, Ballaigues, Switzerland) was used to confirm apical patency. The working length (WL) was established withdrawing 1 mm from the apical foramen. In all experimental groups, a glide path was accomplished by scouting a size #15 K-file (Dentsply Maillefer) up to the WL. Aiming to simulate a closed-ended system, the root apex of each tooth was sealed with hot glue and then embedded in polyvinyl siloxane, promoting air entrapment at the apical region [15].

For the mechanical preparation of specimens, Reciproc R25 and R40 instruments (VDW, Munich, Germany) were used according to the manufacturer's instructions, powered by an electric motor (VDW Silver; VDW). Briefly, R25 file was directed to apical region until the WL was reached. During this procedure, the instrument was used in a reciprocating motion ("RECIPROC ALL") through slight apical pressure and a slow in-and-out pecking motion with an amplitude of approximately 3 mm. After that, R40 instrument was used in the same manner up to the WL. Each instrument was used in one tooth and discarded.

Irrigation was performed with a 31-gauge NaviTip double sideport needle (Ultradent Products Inc., South Jordan, UT, USA), inserted up to 1 mm from the WL, using a total volume of 20 mL of 5.25% sodium hypochlorite (NaOCl) per canal. The specimens were submitted to a new scan and reconstruction procedures applying the aforementioned parameters.

Final irrigation protocols

After the completion of canal preparations, root canals of each group were submitted to a final rinse using 20 mL of solution (16 mL of 5.25% NaOCl and 4 mL of 17% EDTA) in a total time of 5 min according to one of the four final irrigation protocols (Fig. 1):

PUI group. PUI was performed using an ultrasonic tip (20/0.01) (Irrisonic; Helse Dental Technology, São Paulo, Brazil) set at a low power setting (10%) mounted in a piezoelectric ultrasonic device (Enac-Osada, Tokyo, Japan). A 31-gauge NaviTip double sideport needle was inserted 1 mm from the WL in order to flush 4 mL of 5.25% NaOCl. Then, PUI was used in cycles by an intermittent approach. Firstly, the ultrasonic tip was placed 1 mm short of the WL and activated during 30 s. This cycle was repeated twice. Then, another cycle was performed using 4 mL of 17% EDTA. Lastly, one more cycle with 4 mL of 5.25% NaOCl solution was performed.

EndoVac group. Firstly, macroirrigation was accomplished with 6 mL of 5.25% NaOCl for 30 s by the use of EndoVac macrocannula. For this, the tip was inserted into the root canal until finding resistance and moved up and down. Then, root canal space was left with this solution without interferences during 60 s. Following this, 3 cycles of microirrigation succeeded. For the first cycle, the microcannula was inserted 1 mm short of the WL and maintained during 60 s. Simultaneously, 5 mL of 5.25% NaOCl was continuously restocked. Then, microcannula was removed so that the solution was kept without interferences during 60 s. The two successors' microirrigation cycles were similar, but using 4 mL of 17% EDTA and lastly 5 mL of 5.25% NaOCl.

SAF group. A 2.0-mm-diameter SAF instrument (ReDent-Nova) was connected to an RDT3 head (ReDent-Nova) adapted to a vibrating handpiece (GentlePower Lux 20LP; KaVo, Biberach, Germany). The instrument was inserted 1 mm from the WL and operated with an in-and-out motion. A continuous irrigation of 12 mL of 5.25% NaOCl with a 4 mL/min flow rate was applied in each specimen during preparation. After that, the smear layer removal was obtained by the use of 4 mL of 17% EDTA during 1 min. Then, a final flush with 4 mL of 5.25% NaOCl was applied.

EasyClean group. Initially, a 31-gauge NaviTip double sideport needle was inserted 1 mm from the WL in order to flush 4 mL of 5.25% NaOCl. Then, EasyClean instrument was activated 1 mm short of the WL in a reciprocation motion ("RECIPROC ALL") during 30 s. This cycle was repeated twice. Another cycle using 4 mL of 17% EDTA was performed followed by a final cycle using 4 mL of 5.25% NaOCl.

An experienced operator conducted all experimental procedures. Finally, absorbent paper points (Dentsply Maillefer) were used to dry the root canals. Finishing this stage, the specimens were submitted to a postoperative scan and reconstruction with the previously mentioned parameters.

Micro-CT evaluation and quantitative three-dimensional analysis

Specimen image stacks obtained after the tested final irrigation protocols were rendered and co-registered with their respective preoperative datasets by the use of an affine algorithm of the 3D Slicer 4.6.0 software (http://www.slicer.org) [16]. As previously described [3, 17, 18], the quantification of the removal of AHTD was represented for each specimen as the percentage of the total root canal volume after preparation.

Statistical analysis

Initial and final volume (mm^3), surface area (mm^2), and hard-tissue debris accumulation (%) of the final irrigation protocols were calculated as reference parameters to verify if specimens within groups had similar conditions. One-way analysis of variance was applied for the comparison of these parameters between groups. Raw data normal distribution was confirmed using the Shapiro-Wilk test ($P > 0.05$). Post hoc was performed using the Tukey test for multiple comparisons. The alpha-type error was set at 0.05. SPSS 11.0 (SPSS for Windows v17.0; SPSS Inc., Chicago, IL, USA) was used as an analytical tool.

Results

The degree of homogeneity (baseline) of groups was confirmed in relation to canal length, volume, and surface area, as well as the volume of AHTD obtained after canal preparation ($P > 0.05$) (Table 1). All groups presented a decrease on the accumulation of

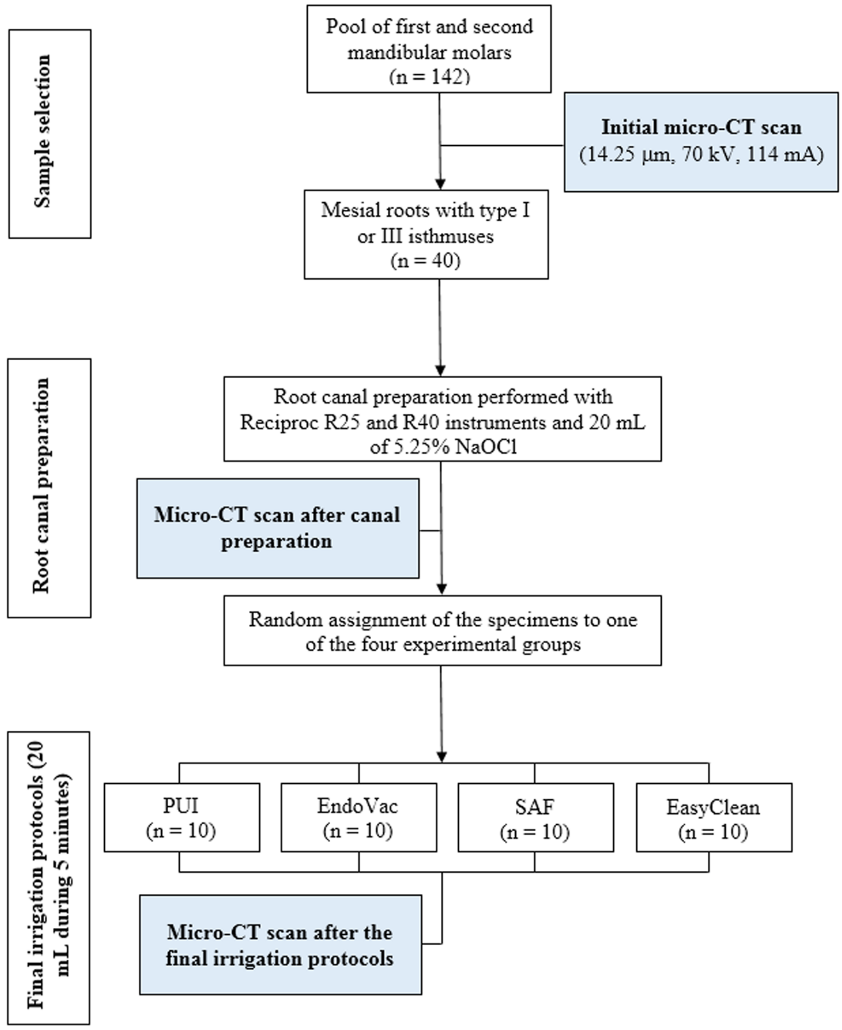

Fig. 1 A flowchart of the experimental procedures

hard-tissue debris after the use of the final irrigation protocols ($P < 0.05$). No significant differences in the removal of AHTD were observed among the final irrigation protocols ($P > 0.05$) (Fig. 2).

Table 1 Mean ± standard deviation of volume and area of the root canal space and hard-tissue debris accumulation according to the preparation stage in each group

Parameters		Preparation stage	Final irrigation protocols			
			PUI	EndoVac	SAF	EasyClean
Root canal space	Volume (mm^3)	Before preparation	5.54 ± 2.54	4.07 ± 1.36	5.60 ± 3.51	3.91 ± 1.86
		After preparation	9.23 ± 2.54	7.95 ± 2.55	11.1 ± 3.37	8.59 ± 2.03
	Area (mm^2)	Before preparation	64.1 ± 18.0	55.4 ± 11.6	70.8 ± 24.9	52.3 ± 17.2
		After preparation	73.9 ± 18.33	66.9 ± 14.6	85.0 ± 22.0	67.7 ± 15.9
Hard-tissue debris accumulation	Volume (mm^3)	After preparation	0.031 ± 0.06	0.027 ± 0.03	0.031 ± 0.05	0.028 ± 0.03
		After final irrigation	0.007 ± 0.01	0.009 ± 0.01	0.019 ± 0.03	0.015 ± 0.02
	Volume (%)	After preparation	0.63 ± 1.56	0.46 ± 0.73	0.32 ± 0.55	0.30 ± 0.34
		After final irrigation	0.14 ± 0.38	0.14 ± 0.27	0.20 ± 0.40	0.17 ± 0.28

Volume (%)—percentage of the hard-tissue debris volume as regards to the root canal volume after preparation

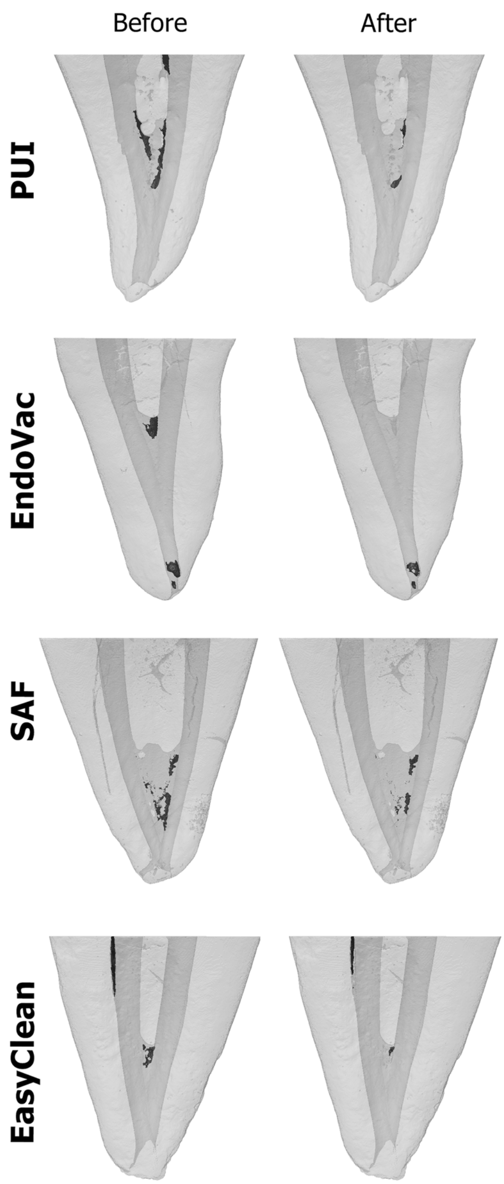

Fig. 2 Representative three-dimensional models of the mesial root canals of mandibular molars before and after final irrigations protocols with PUI, EndoVac, SAF, and EasyClean showing the location of debris (black)

Discussion

The present study assessed the impact of four final irrigation protocols (PUI, EndoVac, SAF, and EasyClean) on the removal of AHTD from isthmus-containing mesial roots of mandibular molars through micro-CT analysis. This nondestructive imaging technology has demonstrated to be the gold standard method for the evaluation of AHTD into the irregularities of the root canal system because it permits a longitudinal observation of the same specimen during several experimental procedures at different time points [2–5, 18].

Isthmus-containing mesial roots of mandibular molars were selected for this study not only because of their high prevalence [19], but mainly as a result of the major challenge that these teeth represent for proper cleaning and disinfection [20–22]. Due to the intrinsic heterogeneity of root canals morphology, known as biological bias, efforts were undertaken to ensure equivalent comparability of groups as regards to the root canal anatomy. For that, a micro-CT pre-screening of specimens based on the configuration and morphology (length, volume, and surface area) permitted to distribute four equated specimen into one of the four experimental groups [5, 23, 24]. As a result, statistics demonstrated an adequate balance between the specimens of groups with respect to canal volume and surface area. This allowed to increase the internal validity of the current study and substantially reduced anatomical biases that may mislead the outcomes.

Besides anatomy, it is important to standardize further experimental conditions as regards to the comparison of final irrigation protocols during the removal of AHTD. Therefore, the ultrasonic tip of the PUI group was used 1 mm short of the WL in order to standardize the depth penetration of the final irrigation protocols. Furthermore, root canal preparation was performed up to Reciproc R40 instrument. According to the manufacturers of Reciproc system, if the root canal is considered narrow, as most of the mesial canals of mandibular molars, the instrument of choice is the Reciproc R25. However, clinically speaking, it is of note that there are no scientifically established guidelines to set the optimal final size of canal preparation. Consequently, different philosophies regarding the optimal canal preparation size and shape are proposed, resulting in some controversy as to whether apical enlargement is indeed necessary. Root canal preparation was performed up to Reciproc R40 because larger apical diameters have been encouraged by the accumulated body of evidence, which indicates that this approach is able to promote a more effective irrigation in the apical region, as well as improve infection control and overall root filling quality [25–27].

In the present study, no significant differences were observed among final irrigation protocols as regards the removal of AHTD ($P > 0.05$). Therefore, the null hypothesis tested was accepted. The present results are in accordance with previous studies that also showed similar debris removal between PUI and EndoVac [28, 29], EndoVac, and SAF [30], as well as PUI and SAF [31].

Although these final irrigation protocols present several differences such as irrigant delivery/aspiration method, activation technique, power setting equipment and kinematic, flexibility, cross-sectional design, core size, and taper, a similar mean percentage removal of debris was encountered. Therefore, despite these considerable differences, all systems seem to be individually effective in increasing the flux and reach of irrigants on isthmus walls, thus resulting in equivalent cleaning of hard-tissue debris. As previously stated, in order to create a reliable comparison between groups, all protocols were submitted to similar irrigation parameters. Consequently, the results may also be explained by the interplay among two factors that allow satisfactory removal of debris during root canal preparation: (i) a high volume (20 mL) of irrigant delivered by these final irrigation protocols [29, 32, 33] and (ii) an apical enlargement of the canals up to a size #40, which allows a higher volume of irrigant reaching the apical area [3, 34]. A previous micro-CT study already showed that the increase of apical preparation size significantly reduced the overall amount of packed debris [3].

The newly developed EasyClean instrument presented a similar performance when compared to PUI, which contrasted with previous findings that showed that the removal of debris was more effective by the former [12] or the latter [35]. These contradictory results may be explained by differences in the methodological design. Kato and co-authors [12] performed the canal preparations up to a size #30 and Prado et al. [35] up to a size #25, while the mesial canals of mandibular molars were instrumented up to a size #40 herein. It was already stated that the increase of apical preparation size plays an important role on the removal of debris [3]. These studies analyzed the removal of AHTD by root levels, while the entire canal was analyzed in this study. In addition, it is important to emphasize that both studies evaluated its efficiency on the removal of AHTD by means of qualitative score-based scanning electron microscopy. In the current investigation, the assessment of the removal of AHTD promoted by the final irrigation protocols was performed through micro-CT imaging. The association of this technology with image analysis permits to automatically extract quantitative data of dentin morphology, reducing human biases [36]. To the best of the authors' knowledge, this is the first study evaluating the removal of AHTD by EasyClean using this methodology.

Although the final irrigation protocols showed a significant decrease in hard-tissue debris accumulation ($P < 0.05$), which can be translated into improved cleanliness ability of the root canal system, none of them was able to render mesial root canals of mandibular molars completely free from dentin particles. This finding is in accordance with several previous studies [3, 28–31, 37, 38], and it underscores that mechanical preparation of root canals invariably creates dense accumulation of debris in areas of anatomic irregularities that cannot be removed with the current available techniques. Thus, this reveals the existing need of the development of new protocols and instruments capable to optimize and enhance the cleaning of root canal complexities. Further researches are necessary to evaluate the impact of this improved cleanliness of the root canal system performed by these final irrigation protocols on the clinical success rate of endodontic treatment.

Conclusions

Under the conditions of this study, it can be concluded that the final irrigation protocols showed the same effectiveness in the removal of AHTD. However, none of them was able to render mesial canals of mandibular molars completely free from packed debris.

Acknowledgements The authors deny any conflicts of interest related to this study. This study was partially funded by FAPERJ and CNPq.

Funding This study was partially funded by FAPERJ.

Compliance with ethical standards

Conflict of interest Emmanuel João Nogueira Leal Silva, Carla Rodrigues Carvalho, Felipe Gonçalves Belladonna, Marina Carvalho Prado, Ricardo Tadeu Lopes, Gustavo De-Deus, and Edson Jorge Lima Moreira declare that they have no conflict of interest.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent For this type of study, formal consent is not required.

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