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Irrigant Agitation Techniques versus Passive Ultrasonic Irrigation for Removing Debris from Curved Root Canals: An Environmental Scanning Electron Microscopic Study

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ABSTRACT

Introduction: The aim of this study was to compare the efficacy of passive ultrasonic irrigation (PUI) *versus* mechanical agitation of the irrigant promoted by the EasyClean (EC) and XP-Endo Finisher (XP-F) systems in removing debris from root canal walls, using environmental scanning electron microscopy (ESEM). **Methods and Materials:** Twelve curved mesiobuccal canals of mandibular molars were prepared with the ProTaper Next system up to file X2 (25/0.06). The specimens were embedded in flasks containing silicone putty, sectioned lengthwise, and a 4-mm long groove was made on the canal wall of the buccal half of the specimen, extending from 2 mm up to 6 mm short of the apex. Five cross-sectional markings were made along this groove to establish standardized locations for imaging. The same specimens were used to prepare a negative control group (without debris), a positive control group (completely covered by debris), and 3 experimental groups according to the final irrigation protocol employed: PUI, EC or XP-F. ESEM images were obtained and evaluated by 3 examiners. The amount of debris observed on the images was classified according to a 4-category scoring system. The kappa test was used to assess inter-examiner agreement, and the Kruskal-Wallis and Dunn tests were used to compare the scores ($P<0.05$). **Results:** The scores attributed to the PUI, EC, and XP-F groups were statistically similar to those attributed to the negative control group ($P>0.05$). **Conclusion:** Based on this *in-vitro* study, the mechanical agitation of the irrigant promoted by EC and XP-F was as effective as using PUI to remove debris from the root canal walls.

Keywords: Debridement; Dental Instruments; Root Canal Irrigants; Scanning Electron Microscopy; Ultrasonic Surgical Procedures

Introduction

The anatomical complexity of the root canal system (RCS) favors the permanence of tissue and microbial remnants, even when instrumentation is performed carefully [1], and these may compromise the disinfection process [2, 3]. In addition, the instrumentation procedure itself produces a residual layer of contaminated organic and inorganic debris adhered to the dentinal surface, and this renders effective cleaning and disinfection of the root canal even more difficult to achieve, ultimately jeopardizing the outcome of endodontic treatment [4]. Irrigation plays a critical role in this scenario by acting on areas that are difficult to reach by instrumentation [5, 6], particularly in the apical region [7].

The irrigant reduces the amount of dentin debris, not only due to its chemical effect, but also to the mechanical action used to deliver it [8]; however, scientific evidence suggests that the cleaning action of conventional irrigation is limited [9-11].

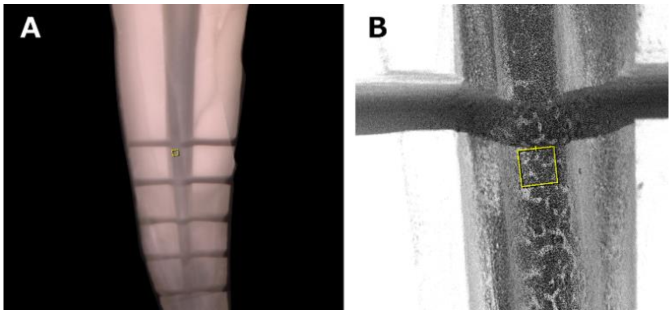

Figure 1. *A*) Specimen illustrating the longitudinal groove created in one of the root canal walls and the 5 cross-sectional markings cut into the root dentin at every millimeter; *B*) To establish standardized apical locations to be examined using ESEM imaging

Thus, the search for irrigation agitation techniques to optimize cleaning and disinfection, especially in confined areas, constitutes a broad line of research in contemporary endodontics [2, 8, 12, 13].

Passive ultrasonic irrigation (PUI) is one of the most cited techniques in the literature [13-15]. It is based on the production of the cavitation and acoustic microstreaming phenomena to enhance the cleaning action of the irrigant [14]. However, the reach of this technique is limited in curved canals [10, 15, 16], because the ultrasound instrument is less likely to oscillate freely.

The EasyClean (EC) mechanical agitation system (Easy Equipamentos Odontológicos, Belo Horizonte, MG, Brazil) consists of an instrument made with acrylonitrile butadiene styrene (ABS). Supplied in size 25/.04 and having a working portion shaped like an aircraft wing, the instrument has proved effective and safe for cleaning the final portion of curved roots, considering that its flexibility and lack of cutting blades rule out the risk of producing deformities on the canal walls [17].

The XP-Endo Finisher (XP-F) instrument (XP-F; FKG Dentaire, La Chaux-de-Fonds, Switzerland) comes in size #25, has no taper, and is produced in a NiTi alloy denominated Max Wire. This alloy undergoes a molecular phase change at temperatures above 35°C. Thus, the instrument is straight when predominantly in the martensitic phase, and takes on a semi-circumferential shape by influence of the austenitic phase, thereby enhancing its ability to adapt three-dimensionally to RCS walls [18, 19]. Furthermore, its design and elliptical movement inside the canal can further contribute to its reaching inaccessible areas of the canal [20].

The aim of this *ex-vivo* study was to conduct an environmental scanning electron microscopy (ESEM) comparison of the efficacy of PUI *versus* that of the mechanical activation of the irrigant produced by the EC and XP-F systems in removing debris from root canal walls at 5 pre-established root levels. The null hypothesis was that there would be no significant difference among the final irrigation techniques tested.

Materials and Methods

This study was approved by the local research ethics committee (register no. 2.379.996). The study specimens were 12 mesial roots of human mandibular molars, donated expressly by patients whose teeth were indicated for extraction. The sample size was calculated using R software (Bell Laboratories, Lucent Technologies, Murray Hill, NJ, USA), considering a test power of 80% and a type I error probability of 0.05.

Teeth that had fully formed roots, whose mesial canals had independent foramina, curvatures between 15° and 20° [21], and an initial foramen diameter corresponding to a #10 K-type file (Dentsply Sirona Endodontics, Ballaigues, Switzerland) were included. Teeth that had calcifications or root resorptions, or that had been submitted to previous endodontic treatment were excluded.

After gaining coronal access, a #10 K-type file (Dentsply Sirona Endodontics) was inserted into the mesiobuccal canal using oscillatory movements, until its tip was visible at the apical foramen. A silicone stop was fitted at the tip of the corresponding cusp to obtain the initial measurement of the specimen. The occlusal surface was abraded to standardize the specimen length at 19.0 mm, using a diamond disk (Horico Dental Hopf, Berlin, Germany) coupled to a handpiece and low-speed micro-motor under refrigeration. The working length (WL) was established 1 mm short of the apical foramen.

The distal root of the specimen was sectioned with a diamond disk (Horico Dental Hopf) under refrigeration, and then discarded. The specimen was secured to a bench vise, and the mesiobuccal canal was instrumented by a single operator using instruments X1 (17/0.04) and X2 (25/0.06) of the ProTaper Next system (Dentsply Sirona Endodontics). The instruments were taken up to the WL using in-and-out movements, and a brushing motion was applied when removing them from the canal. They were driven by an X-Smart Plus motor (Dentsply Sirona Endodontics), set to operate at 300 rpm and 2 N.cm torque, using continuous rotation.

Foraminal patency was confirmed at each instrument change with a #10 K-type file (Dentsply Sirona Endodontics), and irrigation was performed with a 2.5% sodium hypochlorite (NaOCl) solution delivered with a hypodermic syringe and 30-gauge needle (Ultradent, South Jordan, UT, USA), using a total volume of 20 mL per specimen.

A longitudinal groove was created on the mesial and distal aspects of the specimen, along the entire length of the root, and deepened into the dentin leading up to the canal, but without reaching it. This procedure was performed with a 0.08 mm thick diamond disk (Horico Dental Hopf) assisted by a dental operating microscope (Stemi 508; Carl Zeiss, Oberkochen,

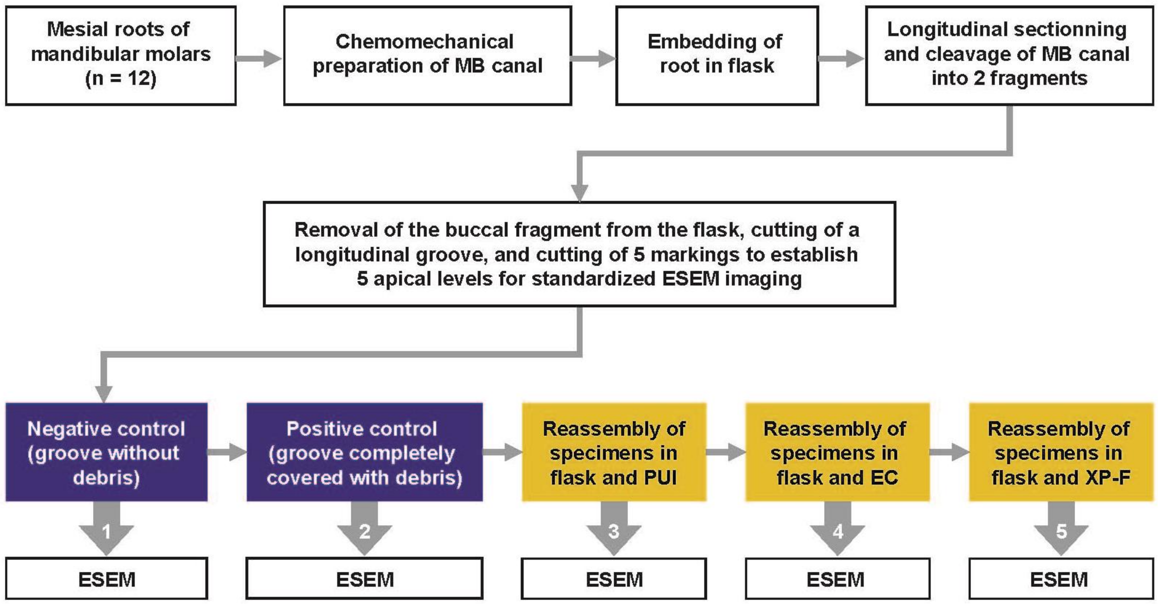

Figure 2. Flowchart demonstrating the experimental procedures performed in the study

Germany) under 8× magnification. The roots were then washed in running water to remove debris.

The root surfaces were abraded using a diamond disk (Horico Dental Hopf; Ringleb, Berlin, Germany) under constant irrigation to render them thinner, thereby reducing specimen moisture and possible interference of this factor with the ESEM imaging procedure [16]. The roots were then embedded up to the cementoenamel junction in flasks containing silicone putty (Elite HD; Zhemarck, Badia Polesine, Italy). After letting the silicone set, the specimens were cleaved in half with a #24 spatula. The buccal part of the specimen was removed using hemostatic forceps, and a standard longitudinal groove measuring 4.0 mm long, 0.2 mm wide and 0.5 mm deep was made on one of the canal walls, extending from 2 mm up to 6 mm short of the apex, following the method proposed by Van der Sluis [14]. Cross-sectional markings were made at each millimeter along the groove, totaling 5 markings, in order to establish standardized locations for imaging (Figure 1). The specimens were then washed under running water for 1 min to remove debris.

Control groups

In the negative control group, the specimens were immersed in an ultrasonic bath of a 2.5% NaOCl solution for 3 min, then in a 17% ethylenediaminetetraacetic acid (EDTA) solution for another 3 min, and then washed in distilled water for 1 min. Afterwards, they were oven-dried at 80°C for 3 min and analyzed using ESEM, under 1.750× magnification. Images were obtained of the 5 pre-established locations along the prepared groove.

In the positive control group, dentin from the previously discarded root was abraded with a low-speed #2 spherical bur (KG Sorensen, Cotia, SP, Brazil), collected, and stored in a 2.5% NaOCl solution for 5 min. This dentin was then smeared on the external and internal surfaces of the groove with a brush (Endo Tim; Voco, Cushaven, Germany), simulating a situation of debris accumulation in areas of irregularities. The specimens were dried, and new images were obtained following the same procedure performed for the negative control group.

Experimental groups

In the three experimental groups, the specimens were coated with the dentin mass, as performed in the positive control group, and then reassembled in the silicone flasks. The correct fit of the cleaved specimen halves was verified by inserting a gutta-percha cone into the canal and taking digital radiographs in the buccolingual and mesiodistal directions [16, 22].

In the PUI Group, the irrigating solution was delivered at a point 2 mm short of the WL with a 30-gauge needle (Ultradent, South Jordan, Utah, USA), and agitated with an E1 Irrissonic insert (20/0.01; Helsen, Santa Rosa de Viterbo, SP, Brazil) coupled to an ultrasonic unit (ENAC; Osada, Aichi, Japan) set to operate at power level #3. The insert was also positioned 2 mm short of the WL. Five milliliters of 2.5% NaOCl followed by 5 mL of 17% EDTA and then by another 5 mL of 2.5% NaOCl were used. All the solutions were renewed and delivered in 3 cycles of 20 sec, and a final irrigation was performed with 20 mL of distilled water.

In the EC Group, the instrument was inserted up to the WL and driven by the X-Smart Plus motor (Dentsply Sirona Endodontics , Ballaigues, Switzerland), set to operate at 1200 rpm and 2 Ncm torque using continuous rotation. In the XP-F group, the instrument was applied using slow and smooth longitudinal movements extending 7-8 mm to cover the entire WL, and the motor was set to operate at 800 rpm and 1 Ncm torque.

All of the procedures described for the PUI, EC and XP-F experimental groups were performed according to the recommendations of the manufacturers of the devices used for their respective protocols. The same 12 specimens were used in all the study groups, and the application sequence of the irrigants, the disassembly, reassembly and processing of the specimens, and the taking and analysis of the ESEM images were also the same. The experimental procedures performed in the study are illustrated in Figure 2.

Evaluation of the ESEM images

The images were coded according to study group, specimen, and reading site (Figure 3). All of the images from the control and experimental groups for each level were loaded into presentation software (MS PowerPoint, Microsoft Corporation, Redmond, WA, USA) and displayed on an LCD monitor. Three independent examiners, previously calibrated and blind to the study, analyzed and classified the images using a 4-category scoring system adapted from Gambarini and Laszkiewicz [23], as follows:

Score 1: open dentinal tubules with no debris
Score 2: open dentinal tubules without debris in more than 50% of the examined area
Score 3: open dentinal tubules without debris in less than 50% of the examined area
Score 4: dentinal tubules completely covered by debris in 100% of the examined area.

Statistical analysis

The kappa test was used to assess inter-examiner agreement. The Kruskal-Wallis and Dunn tests were used to perform multiple comparisons of the scores attributed to the images. All the statistical calculations were performed using R software (Bell Laboratories, Lucent Technologies, New Jersey, USA). The level of significance adopted was 5%.

Results

The level of agreement among the three examiners was assessed for all of the study groups and root canal levels, and agreement was found to be almost perfect (kappa values ranging from 0.91 to 1.0). The distribution of the scores attributed to the images of the study group specimens at the 5 apical levels examined is shown in Table 1.

The scores attributed to the positive control group were significantly higher than those attributed to the other groups, in all the apical levels analyzed ($P \leq 0.05$). There were no significant differences among the negative control and the experimental groups ($P \geq 0.05$).

Table 1. Mode scores attributed to the images of the specimens according to study group and apical level examined

Group	Pre-established root levels in relation to the apex				
	2 mm	3 mm	4 mm	5 mm	6 mm
NC	Score 1 (100%) [a]	Score 1 (100%) [a]	Score 1 (100%) [a]	Score 1(100%) [a]	Score 1 (100%) [a]
PC	Score 4 (100%) [b]	Score 4 (100%) [b]	Score 4(100%) [b]	Score 4 (100%) [b]	Score 4 (100%) [b]
PUI	Score 1 (70%) [a] Score 2 (30%)	Score 1 (90%) [a] Score 2 (10%)	Score 1 (90%) [a] Score 2 (10%)	Score 1 (100%) [a]	Score 1 (90%) [a] Score 2 (10%)
EC	Score 1 (80%) [a] Score 2 (20%)	Score 1 (80%) [a] Score 2 (20%)	Score 1 (90%) [a] Score 2 (10%)	Score 1 (100%) [a]	Score 1 (90%) [a] Score 2 (10%)
XP-F	Score 1 (70%) [a] Score 2 (30%)	Score 1 (90%) [a] Score 2 (10%)	Score 1 (80%) [a] Score 2 (20%)	Score 1 (100%) [a]	Score 1 (80%) [a] Score 2 (20%)
***P*-value**	<0.001	<0.001	<0.001	<0.001	<0.001

NC, negative control; PC, positive control; PUI, passive ultrasonic irrigation; EC, EasyClean system; and XP-F, XP-Endo Finisher system. Different letters within the column indicate statistically significant differences (Kruskal-Wallis test complemented by the Dunn test, P<0.05)

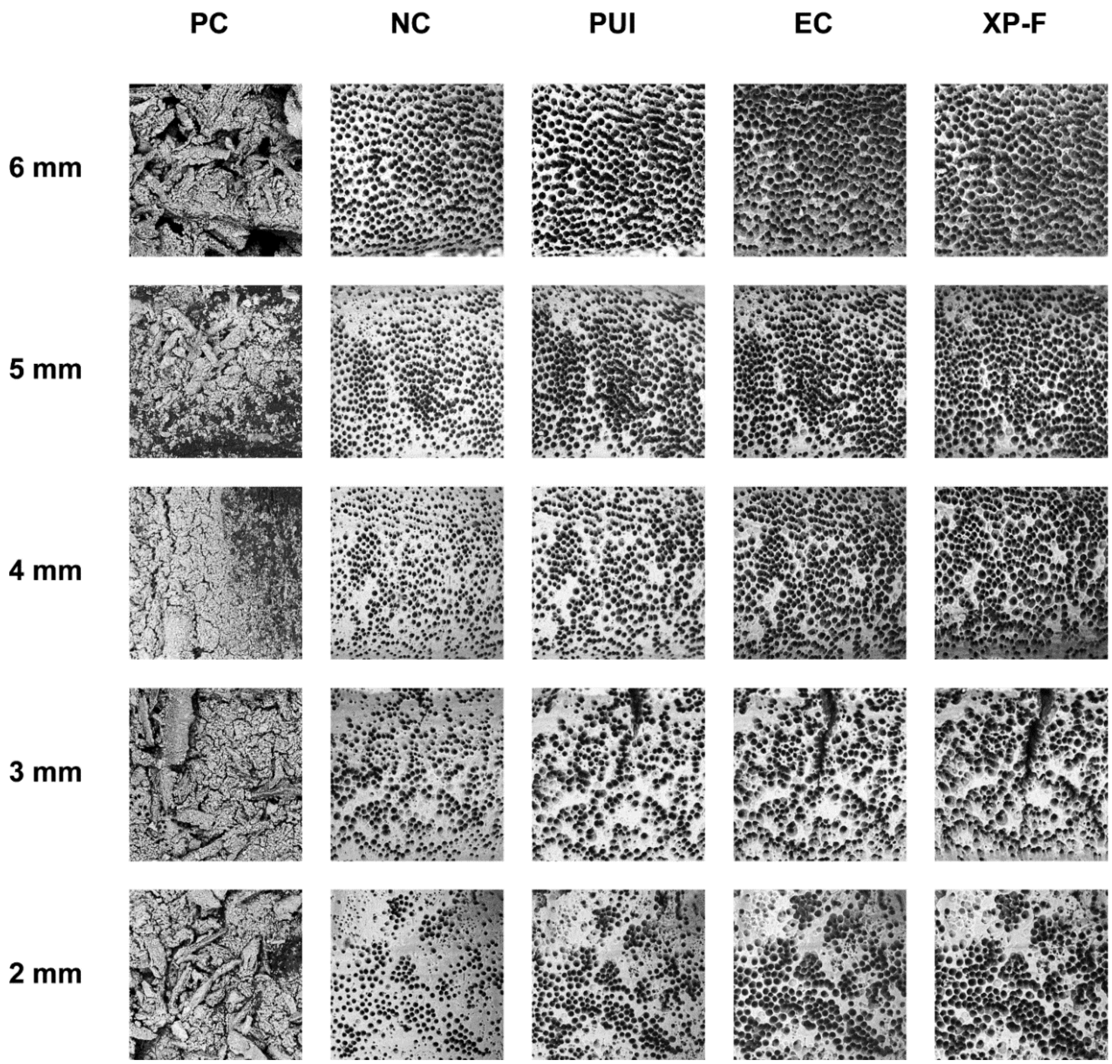

***Figure* 3.** ESEM images representative of groups PC (positive control: completely covered by debris), NC (negative control: without debris), PUI (passive ultrasonic irrigation), EC (EasyClean system), and XP-F (XP-Endo Finisher system), at the five pre-established apical levels (1.750×)

Discussion

There were no significant differences among the mode scores attributed to the negative control and those attributed to the experimental groups, in all of the apical levels analyzed. This demonstrates that the systems tested were equivalent in terms of the degree of final cleanliness that they promoted. Therefore, the null hypothesis was not rejected.

Studies have shown that the apical third is the region with the highest amount of debris adhered to the dentin walls, and irrigant agitation in this region is widely recommended [16, 24-26]. Accordingly, a longitudinal groove was created in the present study, starting at 2 mm and ending at 6 mm from the apex, and was filled with dentinal debris, following previously described methodology [14].

Embedding of the specimens in a flask system enabled reassembling them after cleavage, thus simulating a closed irrigation and aspiration system [24, 27]. Mesiobuccal canals of mandibular molars with moderate curvatures [21] were used because of the frequent occurrence of these conditions in clinical practice. An X2 file of the ProTaper Next system was used to standardize the preparation owing to its size (25/0.06), which allows the safe use of both the EC and XP-F instruments, as well as the application of PUI.

Conventional scanning electron microscopy (SEM) is a widely used method for analyzing root canal cleansing [28, 29]; however, it is impossible to reuse the specimens after the analysis, because of the processing involved, and the sample heterogeneity resulting from having to use different specimens may preclude reliable results [30]. In contrast, the ESEM used in the present study does not require total dehydration and metallization of the specimens, which can be reused, thus eliminating the potential bias caused by anatomical and histological variations of the root dentin [16, 31]. In addition, markings were cut at pre-established levels of the root canal wall to standardize the evaluation sites, thus rendering the assessments less subjective.

PUI is a well-established technique widely reported in the literature, and is considered the gold standard for final endodontic irrigation, as discussed in several studies that confirm its effectiveness and safety in optimizing cleanliness and disinfection [5, 14, 32]. Nevertheless, during the experiment, a deviation occurred in the apical third of one specimen after this technique was performed. This is consistent with previous studies [17, 33, 34] that reported that it was impossible to keep ultrasonic instruments from touching the canal walls during preparation. The purpose of positioning the insert 2.0 mm short of the WL in the present study was to reduce the risk of excessive contact, which could contribute indirectly to the accumulation of debris arising from the activation process itself.

The EC irrigant agitation system was designed to promote the mechanical dragging of the debris adhered to the canal walls when applied up to the WL [16]. In the present experiment, EC was used in continuous rotation, following the methodology used by Andrade-Junior *et al.* [17], although some studies suggest its use in reciprocating movement [16, 31]. One advantage of using continuous rotation is not having to rely on a specific motor capable of providing reciprocating motion, thus favoring universalization of this technique.

The XP-Endo Finisher instrument expands within the root canal at body temperature, and gains access to untouched areas during instrumentation without damaging the dentin or changing the original canal shape [35]. Recent studies have shown that the performance of this system was similar to that of PUI [19, 36], confirming the findings of the present study.

The results of the PUI, EC and XP-F groups were similar to those of the negative control group along the entire length of the examined groove, therefore equally effective in cleaning the root canal walls. This contrasts with the results found by Kato *et al.* [16], who observed that EC was more effective than PUI in the more apical regions of the canal. However, the ultrasonic insert was positioned at the WL in the study by Kato *et al.* [16], whereas it was positioned 2 mm short of the WL in the present study. In addition, the dentin debris was compacted along a longitudinal groove created on one of the canal walls in the present study, whereas indentations perpendicular to the canal wall were created in the study by Kato *et al.* [16], which probably resulted in different contact surfaces. These differences may have contributed to obtaining different results.

The irrigant agitation protocol used in all the groups involved 3 cycles of 20 sec, possibly promoting a cumulative effect in removing the debris [14]. However, a certain degree of erosion was observed on the dentin walls after using the irrigant agitation systems in two specimens. These results are consistent with those of other studies, according to which the alternating use of irrigating agents and the agitation of the solutions may cause dentin erosion [31, 37-39]. Furthermore, reusing the same specimens in the different groups may have contributed to the erosion observed, which may be considered a limitation of the present study.

Conclusions

Based on this *in-vitro* study, the irrigant activation promoted by the EC and XP-F systems was as effective as PUI in removing debris from the root canal walls. Therefore, the EC and XP-F systems can be considered good alternatives to PUI for this step of root canal treatment.

Conflict of Interest: 'None declared'.

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