In Vitro Efficacy of XP-endo Finisher with 2 Different Protocols on Biofilm Removal from Apical Root Canals

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Abstract

Introduction: The purpose of this study was to evaluate the effectiveness of the XP-endo Finisher (XPF; FKG Dentaire SA, La Chaux-de-Fonds, Switzerland) in biofilm removal in comparison with conventional needle irrigation (CNI) and passive ultrasonic irrigation (PUI) using an infected tooth model with an artificial apical groove. **Methods:** Fifty-four extracted human single-rooted premolars were selected. Each tooth was split longitudinally into 2 halves, with a groove made in the apical segment of the canal wall. After growing mixed bacteria biofilm for 4 weeks, the split halves were reassembled and instrumented using Vortex Blue files (Dentsply Tulsa Dental, Tulsa, OK) to size 40/.06. The instrumented teeth were randomly assigned to 6 groups (n = 8) according to the final irrigation protocol. Three different techniques (CNI, PUI, and XPF) were performed each with either continuous irrigation or 3-step irrigation. Scanning electron microscopic images were taken to evaluate the amount of residual biofilm inside and outside the groove. **Results:** Robust growth of biofilm was observed in each canal of the controls after 4 weeks. XPF showed the best biofilm removal efficacy inside and outside the groove followed by PUI and CNI ($P < .05$). The XPF 2 group using the 3-step protocol showed better antibiofilm efficiency than the XPF 1 group with continuous irrigation inside the groove ($P < .05$). **Conclusions:** The XP-endo Finisher, as an irrigation agitation technique, may help to remove biofilm from hard-to-reach areas in the root canal system. The 3-step irrigation protocol was more effective than continuous irrigation when XPF was used. *(J Endod 2017;43:321–325)*

Key Words

Biofilm, conventional needle irrigation, root canal irrigation, ultrasonic, XP-endo Finisher

Significance

The challenge for irrigation may be removal of biofilm from the uninstrumented canal areas. The XP-endo Finisher (XPF) may help to remove biofilm from hard-to-reach areas in the canal system. The 3-step irrigation protocol was more effective than continuous irrigation when the XPF was used.

Anatomic complexities of the root canal system and the presence of microbes as surface adherent biofilm structures serve as the foremost challenges in root canal disinfection (1–4). Even when instrumentation is carefully performed using modern file systems, 30%–50% of the canal wall surface area may remain untouched and covered with biofilm because of irregularities such as fins, isthmuses, ramifications, accessory and lateral canals (2, 5), and the physical constraints of the smaller mesiodistal diameter of the oval canals (6). Therefore, irrigation is of great significance in root canal debridement and also currently the only way to impact the untouched areas (7).

Different irrigating solutions and numerous irrigant delivery devices have been proposed aiming at complete elimination of the microbes from the root canal system (7–9). Sodium hypochlorite (NaOCl) is the most important and widely used chemical irrigant in root canal treatment (7, 10). In addition to the chemical effect, endodontic microorganisms are also reduced by the mechanical action of irrigation (11). The bacteria in biofilm are difficult to eliminate because of their high resistance to treatment (ie, 100- to 1000-fold greater than their planktonic counterparts) (12). Irrigation techniques based on some kind of agitation such as ultrasonics (eg, passive ultrasonic irrigation [PUI]) are found to facilitate irrigant penetration, tissue dissolution, and smear layer removal better than the conventional needle irrigation (CNI) technique (13–15).

A new instrument for irrigant agitation, the XP-endo Finisher (XPF; FKG Dentaire SA, La Chaux-de-Fonds, Switzerland) (16), has been recently introduced as an adjunctive approach to improve the effectiveness of irrigation in endodontics. XPF is an ISO 25/.00 instrument produced using a special type of alloy, the NiTi MaxWire (Martensite-Austenite Electropolish-FleX, FKG). According to the manufacturer, the file is straight in its M phase when it is cooled, and it will change into A phase when it is exposed to body temperature where it will have its unique spoon shape with a length of 10 mm from the tip and a depth of 1.5 mm because of its molecular memory (6, 16). It is suggested to be used at 800 rpm with irrigating solutions after root canal preparation to size #25 or larger.

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Much of the research on endodontic irrigation, from traditional syringe needle delivery to various machine-driven systems (7), has focused on removal of the smear layer (17–20) or hard tissue debris (21, 22). However, an even bigger challenge for irrigation may be the removal of biofilm from the uninstrumented canal areas (7). Therefore, this study was performed to quantitatively analyze the efficacy of 3 different techniques (ie, CNI, PUI, and XPF), each used with 2 different protocols in biofilm removal. A split tooth model and a standardized, multispecies biofilm model with a narrow and deep artificial apical groove and scanning electron microscopy were used. The null hypothesis was that no statistically significant differences would exist among the 6 treatment groups.

Materials and Methods

Standardized Biofilm in a Split Tooth Model

A total of 54 extracted human single-rooted premolars at least 19 mm in length with a closed apex were collected under a protocol approved by the University of British Columbia Clinical Research Ethics Committee Review Boards (certificate H12-02430). The teeth were stored at 4°C in 0.01% NaOCl solution until used.

According to a previously described protocol (23), the teeth were accessed, and the reference cusps were reduced until the length of each tooth was 19 mm, which was determined and measured by inserting a size #10 stainless steel (SS) K-file into the canals. The working length (WL) was set at 18 mm (1 mm short from the apical foramen). To standardize the apical geometry, the canal was hand instrumented to the WL with a size #15 SS K-file followed by ProTaper S1 and S2 rotary NiTi instruments (Dentsply Tulsa Dental, Tulsa, OK). Dentin debris removal must be visible in the apical 4-mm portion of the S2 file.

Two longitudinal grooves were prepared on the buccal and lingual surface of each tooth using a diamond disc (Brasseler, Savannah, GA) under an operating microscope (SZ2-ST; Olympus Corporation, Tokyo, Japan), avoiding penetration into the canal. The tooth was then split into 2 halves with a fine razor blade and a hammer. Only teeth in which the 2 halves could be fully reassembled were included in the study.

A standardized groove of 3-mm long, 0.2-mm wide, and 0.8-mm deep was cut using a small diamond disc (Brasseler) in each canal wall, starting 2 mm from the apex, in order to simulate a deep, narrow fin. Four notches were made at 2, 3, 4, and 5 mm from the apex on each side as reference points to help identify the apical, middle, and coronal thirds of the apical groove after biofilm removal efforts.

The split halves of each tooth were reassembled and fixed with a 5-mm-diameter wax ball over the root tip. Half of the split tooth and wax were placed in a pour of dental stone (Whip Mix, Louisville, KY) using a thin layer of Vaseline (Unilever, Toronto, Canada) as a separator. This was followed by a second pour of dental stone covering the remaining half of the tooth and wax. The apical space left by the wax served as the apical cavity (simulated lesion) in the custom block.

To remove the smear layer, the split halves were opened and rinsed with 5.25% NaOCl for 3 minutes, distilled water for 30 seconds, 17% EDTA for 3 minutes, and then distilled water for 30 seconds twice. Mixed human subgingival plaque bacteria from an adult volunteer were used to grow the biofilm. The split halves were placed in a 12-well plate and incubated in brain-heart infusion (BHI; Becton-Dickinson, Sparks, MD) broth in an anaerobic environment (Bactron300; SHEL LAB, Cornelius, OR) at 37°C for 4 weeks. Fresh BHI broth was changed once a week (23). Earlier studies using the same culture method with BHI have shown growth of several bacterial morphotypes, including spirochetes (12).

Root Canal Instrumentation and Irrigation

After biofilm growth, the split halves were reassembled and placed in the custom blocks. Root canal preparation was performed with a crown-down technique using Vortex Blue (Dentsply Tulsa Dental) in the sequence of #40/.04,#35/.04, #30/.04, and #25/.04 to the WL and then up to #40/.06 as the last file to the WL. The canal was rinsed with 1 mL 3% NaOCl (at room temperature) before each file and after the last file, a total of 6 mL NaOCl per canal. The same experienced operator completed all root canal preparations.

The specimens were then randomly divided into 6 groups ($n = 8$ teeth per group). All canals were irrigated with 3.5 mL 3% NaOCl just before the experimental irrigation/agitation procedures.

Group CNI 1 (Continuous CNI). The canal was flushed with a continuous flow of 1.5 mL 3% NaOCl (90 seconds) using a syringe and a 30-G side-vented needle (ProRinse, Dentsply Tulsa Dental) 1 mm short from the WL.

Group CNI 2 (CNI, 3 Steps). In this group, syringe delivery of 1.5 mL 3% NaOCl was performed 3 times, 0.5 mL 3% NaOCl for 30 seconds each.

Group PUI 1 (Continuous PUI). One milliliter 3%NaOCl was continuously infused into the access cavity and passively activated using an ultrasonic device (ProUltra, Dentsply Tulsa Dental) with an E12 endodontic tip and a U file #20 (NSK Europe GmbH, Eschborn, Germany) at a power setting of 3 for 1 minute simultaneously and then rinsed with 0.5 mL 3% NaOCl for 30 seconds to the WL. The U file was placed 1 mm short from the WL.

Group PUI 2 (PUI, 3 Steps). Passive ultrasonic activation of the irrigant was performed for 20 seconds followed by 0.5 mL 3% NaOCl for 10 seconds to the WL. This 30-second cycle was repeated 3 times.

Group XPF 1 (Continuous XPF). XPF was placed in a contra-angle handpiece (Dentsply Tulsa Dental), inserted into the canal, and then activated (800 rpm and 1-Ncm torque) for 1 minute using slow and gentle 7- to 8-mm lengthwise parietal movements to contact the full length of the canal as suggested by the manufacturer (16). During the procedure, 1 mL 3% NaOCl was continuously supplied to the access cavity. After removing the instrument from the canal while still rotating, the canal was rinsed with 0.5 mL 3% NaOCl for 30 seconds.

Group XPF 2 (XPF, 3 Steps). XPF was used for 20 seconds with 3% NaOCl in the canal followed by 10 seconds of irrigation with 0.5 mL 3% NaOCl. This cycle was repeated 3 times.

In all groups, a final irrigation with 1 mL sterilized water for 30 seconds and 4 mL 17% EDTA for 2 minutes was performed.

Scanning Electron Microscopy

After the instrumentation and irrigation were completed, the teeth were disassembled, and the split halves were fixed in 2% glutaraldehyde, dehydrated in graded series of ethanol solutions, critical point dried, coated with iridium, and examined under scanning electron microscopy (Hitachi SU-3500; Hitachi High-Technologies Inc, Rexdale, Ontario, Canada). To avoid bias in the acquisition of scanning electron microscopic images for evaluation of biofilm removal efficacy, a stratified random sampling was used, which includes predetermining the sampling location from inside the groove (apical, middle, or coronal third) at a low magnification (15×) and then zooming into that area to a higher magnification (1000×) to obtain 3 sample areas (areas in the middle and its adjacent areas on each side) and zooming into the 3 sample areas at 2000× respectively for evaluation. An additional 3 images at 2000× magnification were taken outside the groove in the areas adjacent to the former 3 sample areas inside the groove. For each specimen (1 split half), 3 standardized images at a magnification of

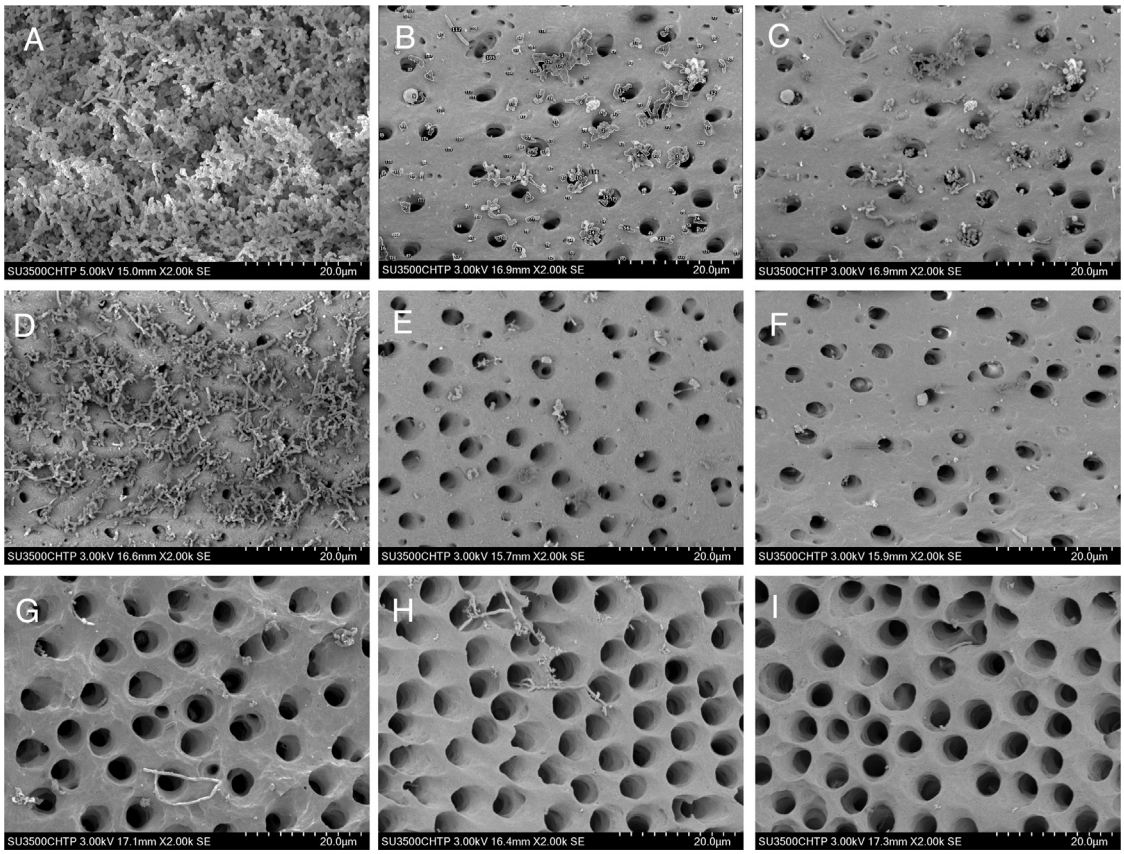

Figure 1. Representative scanning electron microscopic images showing (*A*) the robust growth of multispecies biofilm in a control specimen; (*B*) the measurement of the percent area covered with bacteria semiautomatically using Image-Pro Plus 6.0 software; (*C–F*) the areas inside the groove after irrigation using (*C*) PUI (3 steps), (*D*) CNI (3 steps), (*E*) XPF following the continuous protocol, and (*F*) XPF following the 3-step protocol; and (*G–I*) areas outside the groove after continuous irrigation using (*G*) CNI, (*H*) PUI, and (*I*) XPF.

2000× were obtained from all thirds inside and outside of the groove (a total of 18 images) for analysis. The controls (untreated canals) were examined to ensure the presence of consistent and thick biofilm in the root canal.

Seventeen hundred twenty-eight scanning electron microscopic images acquired from 48 treated teeth were randomly coded and evaluated separately by 2 independent examiners. The percentage of biofilm bacteria remaining was measured using Image-Pro Plus 6.0 (Media Cybernetics, Bethesda, MD) software. The objects were determined semiautomatically using the magic wand function (Fig. 1*A–I*). The examiners can only adjust the software's contrast to discern between the biofilm bacteria and artifacts in the digitized images. Interobserver and intraobserver reproducibility were measured using the weighted coefficient kappa.

Statistical Analysis

Two-way analysis of variance was used to compare the percent area covered with bacteria inside and outside the groove between different treatment groups and different areas within 1 group, with a significance level set at 5%.Statistical analysis was performed with SPSS 16.0 for Windows (SPSS Inc, Chicago, IL).

Results

The interobserver and intraobserver reproducibility were 0.90 and 0.95, respectively. Scanning electron microscopic images taken from inside and outside the groove after treatment using different irrigation techniques and protocols as well as the controls with no treatment are shown in Figure 1. All controls presented robust and consistent growth of multispecies biofilm in the canal under scanning electron microscopic observation after the 4-week incubation period. The average percentage of biofilm remaining from inside and outside the groove of each group is detailed in Table 1.

The 2 XPF groups showed the best biofilm removal efficacy both inside and outside the groove followed by the 2 PUI groups. The 2 CNI groups showed the lowest reduction in surface covered by biofilm ($P < .05$).

Within the groove, group XPF 2 (3-step irrigation) presented better efficiency in biofilm removal than group XPF 1 ($P < .05$). Outside the groove, both XPF groups showed similar effectiveness ($P > .05$). No significant differences were found between PUI 1 and PUI 2 or between CNI 1 and CNI 2, whether inside or outside the groove ($P > .05$).

For all the groups, significantly more bacteria were removed from outside the groove than inside the groove ($P < .05$). Within each group, no statistical differences were found among the apical, middle, and coronal thirds, both inside and outside the apical groove ($P > .05$).

Discussion

In the present study, a multispecies biofilm model using extracted single-rooted teeth with an artificial, standardized groove in the apical canal was used to investigate the effectiveness of 3 different irrigation

TABLE 1. Average Percentages of Bacteria Remaining Inside and Outside the Groove after Irrigation with Conventional Needle Irrigation (CNI), Passive Ultrasonic Irrigation (PUI), and XP-endo Finisher (XPF) Techniques (Mean ± Standard Deviation)

	Inside groove				Outside groove			
Group*	Apical	Middle	Coronal	Total[a]	Apical	Middle	Coronal	Total[b]
CNI 1	25.79 ± 9.22	25.88 ± 7.79	29.33 ± 10.53	27.00 ± 9.33[c]	1.29 ± 1.15	1.69 ± 1.37	1.75 ± 1.61	1.58 ± 1.39[g]
CNI 2	26.46 ± 9.23	25.30 ± 7.48	28.68 ± 8.84	26.81 ± 8.61[c]	1.34 ± 0.95	1.51 ± 0.84	1.75 ± 1.36	1.53 ± 1.08[g]
PUI 1	6.17 ± 3.67	5.92 ± 2.45	5.90 ± 2.52	6.00 ± 2.91[d]	1.27 ± 1.02	1.20 ± 0.93	1.01 ± 0.56	1.16 ± 0.86[f]
PUI 2	5.65 ± 2.96	5.94 ± 2.47	5.57 ± 2.52	5.72 ± 2.65[d]	1.14 ± 0.93	1.28 ± 1.06	0.98 ± 0.36	1.13 ± 0.84[f]
XPF 1	2.95 ± 2.35	2.71 ± 1.98	3.16 ± 2.16	2.94 ± 2.16[e]	0.77 ± 0.44	0.83 ± 0.46	0.85 ± 0.48	0.82 ± 0.46[h]
XPF 2	0.90 ± 0.47	1.12 ± 0.62	1.09 ± 0.63	1.04 ± 0.59[f]	0.74 ± 0.42	0.79 ± 0.40	0.86 ± 0.45	0.80 ± 0.42[h]

Lower case letters indicate statistically significant differences between groups ($P < .05$).

*Protocol 1 in each group was continuous 90 seconds of irrigation with agitation (agitation only with PUI and XPF); protocol 2 was intermittent agitation and irrigation, 3 times 20 + 10 seconds.

techniques used as a continuous or a 3-step protocol in removing the biofilm. This biofilm model is an effort to simulate the complexity of the root canal anatomy as described in a previous study (23). In a previous study by Lin et al (23), the groove was made with a microsurgical blade; in the present study, a small thin diamond disc was used. This allowed easier standardization and increasing the groove depth from 0.3 mm to 0.8 mm. The depth of the groove was partly based on a study by Bellucci and Perrini (24), who reported that the thickness of radicular dentin and cementum in maxillary and mandibular premolar teeth 4 mm coronal to the anatomic apex was 0.95 and 1.05 mm, respectively.

The antibiofilm efficacy of PUI compared with CNI has been debated, with some reports showing enhancement of removal by PUI (25–28), whereas some studies did not find a difference (29–31). In the present study, the PUI groups showed much better biofilm removal than the CNI groups, especially inside the artificial groove. The different findings of the aforementioned studies may be attributed to the differences in the methodological design, differences in root canal anatomy, type of bacteria/biofilm incubated, length of bacterial incubation, instrumentation protocol (apical size and taper), irrigation solution, PUI protocol, and parameters of CNI irrigation (eg, flow rate, needle type, and position). It is also possible that the areas challenging for irrigation in many studies are not available for analysis in samples prepared for microscopic examination. In the present split tooth model, a deep narrow groove in the main canal makes it possible to examine this area in all samples and compare the effectiveness of different irrigation methods in easy and hard-to-reach areas.

In the main canal space (outside the groove), the XPF groups presented the best results in biofilm reduction, 99.18% and 99.2%, compared with 98.84% and 98.87% for PUI and 98.42% and 98.47% for CNI. Although there are statistically significant differences between the 3 irrigation techniques outside the groove, all methods provided good results in this area. The small differences in the easy-to-reach area in the present study may explain the divergent results in earlier studies between PUI and CNI.

So far, there is only 1 published study about the antibiofilm effect of XPF (32). In that study, the XPF showed the greatest bacterial reduction in the main canal space (98.2%) and the highest killing of bacteria in a 50-μm depth (ranging from 78%–82%) into the dentinal tubules compared with standard needle irrigation, use of the EndoActivator (Advanced Endodontics, Santa Barbara, CA), and photon-induced photoacoustic streaming. The study is in line with the results of the present study, which found XPF to be the most effective in removing biofilm from a deep groove of the methods included. However, it should be emphasized that, unlike in the present study, only the main canal with no groove was studied for bacterial reduction in the earlier study.

Inside the groove, XPF provided significantly higher antibiofilm efficiency (97.06% and 98.96%) than PUI (94% and 94.28%) and CNI (73% and 73.19%), which indicates that XPF has excellent potential to eliminate biofilm bacteria from difficult-to-reach areas such as fins. Despite our null hypothesis (no difference), we expected that, if there is a difference, PUI would probably give the best results inside the groove because ultrasonic energy creates both acoustic streaming and cavitation. On the other hand, the effect of cavitation with the ultrasonic file may be limited to just a few micrometers from the file surface (33), and, therefore, only the acoustic streaming could reach the depth of the groove. The size of the groove was so small that the XPF file could not penetrate into it (data not shown), as verified by pilot experiments. Therefore, it is possible that the irrigant flow locally, facilitated by the 800-rpm XP file with an asymmetric structure, created irrigant streaming powerful enough to detach much of the biofilm from the groove.

Interestingly, inside the groove, when the 3-step XPF irrigation technique was performed, less biofilm remained on the dentin surface than with the continuous XPF protocol. This indicates that an intermittent protocol is more effective than continuous irrigation/agitation. This result supports the finding of Druttman and Stock (34), who noticed that the effectiveness of irrigant replacement was increased when the number of cycles of irrigation increased, involving alternate canal agitation using a hand file and syringe irrigation. The possible reason may be that the frequent mixing of the liquid in the canal improved irrigant renewal (14, 34). This means that the irrigation solution containing much debris and biofilm material (eg, loosened by XPF agitation) is brought out, and fresh irrigant has a greater capacity to continue cleaning dissolving and detaching biofilm. Thus, after 3 cycles, a cumulative effect was achieved. Moreover, Macedo et al (35) found that 3 consecutive cycles of refreshment/ultrasonic activation of NaOCl could produce a cumulative increase in the NaOCl reaction rate.

In conclusion, within the limitations of the present study, the XPF, as an adjunctive approach to facilitate irrigation, proved effective in biofilm removal from the main canal as well as from the deep, narrow apical groove. The intermittent, 3-step protocol enhanced cleaning more than the continuous protocol.

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The authors deny any conflicts of interest related to this study.

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