INTRODUCTION

Once the dental pulp becomes irreversibly inflamed or infected, the vital and necrotic remnants of the pulp tissue, debris, microorganisms and microbial toxins need to be eliminated from the root canal system\(^1\). Chemically active irrigants should be included in the treatment regimen\(^2\)–\(^4\) because canal shaping alone cannot debride the whole canal system\(^5\)–\(^6\). The irrigants are required to disinfect and remove debris from the entire canal system which is considered challenging due to the root canal system complexity\(^7\)–\(^8\). Moreover, the formation of apical vapor lock during disinfection might prevent the irrigant from reaching the most apical part of the canal\(^9\). On the other hand, irrigant extrusion beyond the apical foramen into the periapical area is an adverse effect that should be avoided\(^10\)–\(^13\).

Syringe irrigation (SI) has been reported to be ineffective in cleaning the apical region\(^14\),\(^15\) with high risk of irrigant extrusion\(^14\),\(^16\)–\(^19\). Therefore, techniques such as apical negative pressure (ANP) and ultrasonic irrigation have been introduced to prevent these complications and enhance penetration of the irrigant. Placing the ANP aspirating needle at the most apical part displaces the vapour lock, if present, allowing an improved flow of irrigant with better debridement\(^14\),\(^15\),\(^17\),\(^18\) and reducing the apical extrusion of irrigants\(^14\),\(^16\),\(^19\). The EndoVac (EV; Discus Dental, Smart Endodontics, Culver City, CA, USA) uses the ANP concept. It has a master delivery tip, open-ended plastic macrocannula, and closed-ended stainless steel microcannula. The master delivery tip is used to deliver the irrigating solutions in the canal, and the macro- and microcannula are used to aspirate and remove gross and fine debris from the canal, respectively.

The ultrasonically assisted irrigation system has been used to enhance irrigant contact with the canal wall\(^20\) by transmitting acoustic energy from an oscillating needle to the irrigant, resulting in acoustic microstreaming and cavitation\(^20\)–\(^22\). These mechanisms are responsible for a greater reduction of the intracanal biofilm\(^23\),\(^24\). The ultrasonic application mode might be intermittent or continuous. Continuous ultrasonic irrigation (CUI) is delivered via an irrigation source that is connected to the ultrasonic needle. It showed effective irrigation dynamics with continuous replacement of irrigant and keeping the canal filled with irrigant\(^24\). Commercially, the VPro StreamClean system (Vista Dental Products, Racine, WI, USA) is one of the CUI system available with an ultrasonic 30-G needle which is made of nickel-titanium alloy. This system was found to significantly reduce canal debris, compared to the SI\(^17\),\(^25\).

Efforts to maximize root canal cleanliness have been attempted and tested by several endodontic irrigation systems. The effectiveness of the CUI system in endodontic irrigation has not been fully investigated. Thus, the aim of this study was to evaluate the irrigation effectiveness of the CUI, by comparing it to the SI and ANP systems in terms of irrigation extrusion and smear layer (SL) removal. The null hypothesis tested was no difference among the tested irrigation systems in terms of the irrigant extrusion and SL removal.
MATERIALS AND METHODS

Specimen preparation
Forty mandibular anterior incisors with single canals were used in this study. The incisal half of the crown was removed using a low speed saw (Isomet, Buehler, Lake Bluff, IL, USA) to provide straight-line access and a reservoir chamber for the irrigant. The canal length was measured by inserting a #10 K-file (Zipperer, Munich, Germany) into the canal until the tip of the file was observed at the apical foramen and the working length (WL) was determined by subtracting 1 mm from the canal length. Each canal was enlarged to size #35/.04 taper at WL with ProFile rotary files (Dentsply Maillefer, Ballaigues, Switzerland) driven by Dentaport ZX (Morita, Tokyo, Japan). The canals were irrigated with 1 mL of 6% sodium hypochlorite (NaOCl; Vista Dental Products) between files. The patency of each canal was checked by inserting a #10 K-file 1 mm beyond the apical foramen. The root specimens were prepared according to a previous study\(^\text{1}\). Briefly, the root canals were washed with 10 mL of distilled water, dried and then filled with silicone (Examix Fine, GC, Tokyo, Japan) to avoid invasion of saline agar. Each root specimen was then placed inside a flat sided, clear plastic case with warm sterile saline agar and secured by using self-curing resin (Unifast II, GC)\(^\text{1}\). Warm sterile saline agar (BactoTM Agar, Difco, Detroit, MI, USA) colored with 1% acid red (Caries Detector, Kuraray, Osaka, Japan) was placed in the plastic case to evaluate the NaOCl extrusion. Once the agar had set, the silicone material in the canal was removed with an explorer.

Canal irrigation
The root specimens were divided randomly and equally into 4 groups. Groups 1, 2 and 3 had the final irrigation sequence of NaOCl and EDTA solutions performed by using SI, ANP and CUI methods, respectively. Group 4 had the roots ultrasonically irrigated with sterile saline as an irrigant (CUI).

In group 1 (SI), irrigation was performed with a closed end, side-port delivery 27-gauge needle (Vista-Probe\textsuperscript{TM}, Vista Dental Products). The needle was attached to a tubing pump (Masterflex, 7524-40, Cole-Parmer Instrument, Vernon Hills, IL, USA) and inserted 2 mm coronal to the WL or just before binding. An aspiration needle was attached to a suction unit (Minic-W, Shin-ei Industries, Saitama, Japan) and placed at the canal entrance. The irrigation sequence followed was 3 mL of 6% NaOCl, 3 mL of 17% EDTA (Vista Dental Products) and 3 mL of 6% NaOCl.

In group 2, the EV was used according to the manufacturer’s instructions. Constant irrigation with 6% NaOCl for 30 s using the EV delivery/evacuation tip connected to the tubing pump and placed at the canal entrance. The macrocannula was attached to the suction unit and moved up and down from the binding point in the canal to slightly apical to the canal orifice. The canal was left undisturbed, filled with 6% NaOCl for 1 min. Afterwards, 3 cycles of microirrigation were performed with a microcannula attached to the suction unit. In each cycle, the pulp chamber was maintained full of irrigant while the microcannula was placed at WL for 6 s and then moved 2 mm coronally for 6 s, then placed at the WL for 6 s and so forth for 30 s. Then, the microcannula was withdrawn from the canal, which was left filled with irrigant for 1 min. The first and third cycles used 6% NaOCl as an irrigant, while the second cycle used 17% EDTA. At the end of the third cycle, the microcannula was left at the WL to remove excess fluid and the canal was left without replenishment of the irrigant.

In group 3, a constant flow of irrigant was ultrasonically activated with the VPro StreamClean attached to a Piezon Master 400 piezoelectric ultrasonic unit (Electro Medical Systems, Nyon, Switzerland) at medium-low power. The stopper on the needle was set to 2 mm short of the WL. Before activation, the needle was inserted into the canal and the irrigant delivery began. During activation, the needle was slowly moved up and down and constant irrigation with 3 mL of 6% NaOCl, 3 mL of 17% EDTA and 3 mL of 6% NaOCl was delivered.

Group 4 (CUIS) was the same as group 3 except that the final irrigant used was sterile saline.

The total irrigation volume used was 9 mL for all groups. The irrigant was delivered using the tubing pump at a flow rate of 3.0 mL /min. The aspiration pressure of the suction unit was set at ±20 kPa.

Evaluation of irrigant extrusion
After irrigation, all root canals were flushed with 5 mL sterile saline and the plastic cases scanned immediately by an image scanner (GT9600, Seiko Epson, Tokyo, Japan). The images were inspected for discolored areas using Photoshop (version 7.0, Adobe, San Jose, CA, USA) in a 100 mm\(^2\) area (316×316 pixels), the center of which was located at the apex. The discolored areas indicated irrigant extrusion.

Evaluation of SL removal with scanning electron microscope
The root specimens were removed from the plastic cases and the apical 7 mm of each root was sectioned longitudinally and then split into two halves in its buccolingual aspect. They were dehydrated in graded series of ethanol solutions, coated with platinum and observed under a scanning electron microscope (SEM) (S-4500, Hitachi High-Technologies, Tokyo, Japan) at 15 kV. The root canals were photographed at 1,000× magnification to evaluate the presence of SL and observed at 1, 3 and 5 mm from the apical foramen. A 5-level scoring system was employed for assessing the efficacy of SL removal with the criteria proposed by Hülsmann et al.\(^\text{2}\) as follows: 1, no SL, all dentinal tubules open; 2, small amount of SL, some dentinal tubules open; 3, homogeneous SL covering the root canal wall, only few dentinal tubules open; 4, complete root canal wall covered by a homogeneous SL, no open dentinal tubules; and 5, heavy, inhomogeneous SL covering the complete root canal wall. The evaluator was blinded to coded...
RESULTS

The results of the apical extrusion of irrigants are shown in Table 1. The logistic regression model was statistically significant ($\chi^2(2)=6.556$, $p=0.038$). Pairwise comparisons (least significant difference) revealed a significant difference between EV and SI ($p=0.010$) but no significant difference between EV and CUI ($p=0.114$) or CUI and SI ($p=0.317$). Only EV did not extrude irrigant whilst the SI and CUI extruded irrigant in 4 and 2 specimens, respectively (Fig. 1).

SEM smear layer scores of the root canal at the 3 levels are shown in Table 2. There were statistically significant differences between groups at 1 ($p<0.001$) and 3 mm ($p=0.004$). Adjusted $p$ values of the pairwise comparisons are shown in Table 3. No statistically significant differences were found between groups at 5 mm ($p=0.100$).

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Table 1  Extrusion of NaOCl irrigation according to presence or absence of discolored areas

<table>
<thead>
<tr>
<th>Group</th>
<th>No extrusion</th>
<th>Extrusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>SI$^*$</td>
<td>6</td>
<td>4</td>
</tr>
<tr>
<td>EV$^b$</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>VSC$^{a,b}$</td>
<td>8</td>
<td>2</td>
</tr>
<tr>
<td>CUIS</td>
<td>Not Applicable</td>
<td></td>
</tr>
</tbody>
</table>

Different superscript letters indicate statistical difference as revealed by logistic regression analysis.

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Table 2  Distribution of scores for SL removal at 1, 3 and 5 mm after irrigation with different systems

<table>
<thead>
<tr>
<th>Score</th>
<th>SI</th>
<th>EV</th>
<th>VSC</th>
<th>CUIS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 mm</td>
<td>3 mm</td>
<td>5 mm</td>
<td>Total</td>
</tr>
<tr>
<td>1</td>
<td>0</td>
<td>1</td>
<td>3</td>
<td>4</td>
</tr>
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<td>2</td>
<td>0</td>
<td>2</td>
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<td>5</td>
<td>0</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>10</td>
<td>10</td>
<td>10</td>
<td>5</td>
<td>30</td>
</tr>
</tbody>
</table>

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Table 3  Statistical group comparison with $p$ value

<table>
<thead>
<tr>
<th>Group</th>
<th>Comparison</th>
<th>1 mm</th>
<th>3 mm</th>
<th>$p$</th>
</tr>
</thead>
<tbody>
<tr>
<td>EV</td>
<td>VSC</td>
<td>1.000</td>
<td>1.000</td>
<td></td>
</tr>
<tr>
<td>EV</td>
<td>CUIS</td>
<td>0.022*</td>
<td>0.053</td>
<td></td>
</tr>
<tr>
<td>EV</td>
<td>SI</td>
<td>0.001*</td>
<td>0.038*</td>
<td></td>
</tr>
<tr>
<td>VSC</td>
<td>CUIS</td>
<td>0.153</td>
<td>0.086</td>
<td></td>
</tr>
<tr>
<td>VSC</td>
<td>SI</td>
<td>0.010*</td>
<td>0.063</td>
<td></td>
</tr>
<tr>
<td>CUIS</td>
<td>SI</td>
<td>1.000</td>
<td>1.000</td>
<td></td>
</tr>
</tbody>
</table>

*Statistical significance

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Fig. 1  Representative images for irrigant extrusion of SI (a), CUI (b) and EV (c).
**DISCUSSION**

Disinfecting the entire root canal system by irrigation is a prerequisite for a favorable treatment outcome. However, irrigating the most apical canal poses a challenge in attaining cleanliness whilst avoiding irrigant extrusion beyond the apex. The present study used a similar set-up to previous studies, which consisted of an extracted human tooth with its root submerged in a gel that simulated the periapical tissues, and therefore created a closed system. However, it is not known if the resistance exerted by the gel is larger, greater or equal than that exerted by the periapical tissues *in vivo*. Therefore, whether this method produces an over- or underestimation of the actual clinical situation remains to be studied. Nonetheless, the specimens were affected equally. Another method of evaluating irrigant extrusion is by measuring the volume of irrigant extruded from the canal. However, neither method addresses a potential confounder variable that is the influence of the apical anatomy on the potential extrusion of the irrigating solution. The apical third of the root has reportedly the highest incidence of accessory canals and apical deltas. In addition, accessory apical foramina have also been shown to exist in mandibular molars and could very well be present in other teeth. These could be potential portals of exit for the irrigant solution in addition to the main apical foramen and the inability to include this variable in a standardized manner using extracted human teeth represents a limitation of the presently used methods.

In the present *in vitro* study, a comparison between SI, EV and CUI was conducted to investigate the extent of their cleaning efficacy along with the risk of irrigant extrusion in mature permanent teeth. Results detected differences among the tested groups. In term of irrigant extrusion, it was observed in 4 and 2 specimens in SI and CUI, whilst the EV specimens had no extrusion detected (Table 1). In terms of cleaning efficacy, another group (CUIS) was added for comparison where the CUI was utilized with sterile saline as a final irrigant. All groups had comparable cleaning efficacy at 5 mm. EV was able to remove SL significantly more than SI at 1 and 3 mm levels. The CUI had significantly removed more SL than SI at 1 mm.

Mitchell *et al.* reported that increasing the apical diameter increased the likelihood that irrigants are extruded beyond the apex. Therefore, the apical canal preparation was standardized to the minimal size (size 35) that can accommodate the microcannula in EV. Furthermore, the amount of irrigant delivered was controlled to allow for a direct comparison between the three methods. The teeth were embedded in gel creating a closed system to simulate *in vivo* conditions.

Preventing irrigant extrusion is of utmost importance in order to keep the periapical area healthy and avoid any complications. Although the recommended side-vented needle was placed away from the apex, extrusion occurred in 4 specimens. This can be attributed to the use of positive pressure during irrigant delivery. Mitchell *et al.* reported by using complete mature roots in a similar set-up, consistently found a lower occurrence of extrusion for EV compared to the SI. On the contrary, Jamleh *et al.* compared two different ANP systems: EV and intracanal negative pressure (iNP), and found the EV was unable to prevent extrusion in any tooth. This might be attributed to methodology differences since simulated immature teeth was used requiring more caution. Thus, the EV might be used with safety in teeth with complete root formation. Furthermore, continuous ultrasonic irrigation was able to reach the most apical area with a low risk of irrigant extrusion which is in agreement with a previous study.

Clinically, removal of the resultant SL from canal shaping is necessary as it might harbor packed microbes and infected debris in the canal wall. The SI was unable to clean the root canal system effectively, which is consistent to past studies. The EV achieved the best results in terms of SL removal. However, it failed to completely clean all specimens. Ahmetoglu *et al.* reported the same finding. This might be attributed to the sole effect of the chemical means on the SL with short irrigation contact time. The ultrasonic activation was proposed and tested as a mechanical means to agitate the irrigant in order to enhance SL disturbance. Spoleti *et al.* found that, with sterile saline, the ultrasonic activation was able to disinfect the root canal more effectively than the SI. Although the CUIS (CUI with saline) was able to completely remove the SL at the coronal level in 5 specimens that might be due to the ultrasonication, it was unable to remove the SL more apically. Previous studies revealed that ultrasonic irrigation cannot get through the apical area due to vapor lock. However, with the aid of NaOCl, results showed the maximum SL removal in the apical third was attained equally with the EV and CUI (Table 3). Therefore, combining the effect of chemical disinfection, ANP along with mechanical debridement would give much improved cleanliness at the most apical areas.

Within the limitations of this *in vitro* study, differences between the experimental groups were found in terms of irrigant extrusion and SL removal; therefore, the null hypothesis was rejected. The CUI might clean the root canal system effectively; however, it was unable to avoid irrigant extrusion.

**ACKNOWLEDGMENT**

The authors deny any conflict of interests.

**REFERENCES**

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