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Multivariate analysis of the cleaning efficacy of different final irrigation techniques in the canal and isthmus of mandibular posterior teeth

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Objectives: The aim of this study was to compare the cleaning efficacy of different final irrigation regimens in canal and isthmus of mandibular molars, and to evaluate the influence of related variables on cleaning efficacy of the irrigation systems. ***Materials and Methods:*** Mesial root canals from 60 mandibular molars were prepared and divided into 4 experimental groups according to the final irrigation technique: Group C, syringe irrigation; Group U, ultrasonics activation; Group SC, VPro StreamClean irrigation; Group EV, EndoVac irrigation. Cross-sections at 1, 3 and 5 mm levels from the apex were examined to calculate remaining debris area in the canal and isthmus spaces. Statistical analysis was completed by using Kruskal-Wallis test and Mann-Whitney U test for comparison among groups, and multivariate linear analysis to identify the significant variables (regular replenishment of irrigant, vapor lock management, and ultrasonic activation of irrigant) affecting the cleaning efficacy of the experimental groups. ***Results:*** Group SC and EV showed significantly higher canal cleanliness values than group C and U at 1 mm level ($p < 0.05$), and higher isthmus cleanliness values than group U at 3 mm and all levels of group C ($p < 0.05$). Multivariate linear regression analysis demonstrated that all variables had independent positive correlation at 1 mm level of canal and at all levels of isthmus with statistical significances. ***Conclusions:*** Both VPro StreamClean and EndoVac system showed favorable result as final irrigation regimens for cleaning debris in the complicated root canal system having curved canal and/or isthmus. The debridement of the isthmi significantly depends on the variables rather than the canals. (*Restor Dent Endod* 2013;38(3):154-159)

Key words: EndoVac; Isthmus; Ultrasonics; VPro StreamClean

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Introduction

Cleaning and shaping of the root canal system by chemo-mechanical instrumentation and irrigation is an essential procedure for the successful endodontic treatment.[1] Since instrumentation alone cannot provide thorough disinfection of the root canal and might leave untouched area, the importance of antiseptic irrigants and the flushing action of irrigation methods have been emphasized.

There have been many remarkable advances in endodontic irrigation technique to overcome limitations of conventional needle syringe irrigation. The EndoVac system (Discus Dental, Culver City, CA, USA), which applies apical negative pressure to full working length, has been shown to provide more safe and efficient cleansing of the apical third of the root canal.[2-5] Recently introduced VPro StreamClean (Vista Dental

Products, Racine, WI, USA) ultrasonically activates continuously refreshing irrigants through a 30 gauge side vent nickel-titanium needle. According to the studies which investigated the flushing action and disinfection of these new irrigation techniques, the effectiveness of certain irrigation regimen seems to be influenced by physical factors and variables.[6-12] Among them, the dominant factors influencing the efficacy of irrigation can be vastly categorized as delivery mode of irrigation, vapor lock management and ultrasonic activation. In respect of the irrigant delivery mode, Hockett *et al.* reported that negative pressure delivery of irrigants demonstrated better microbial control than positive pressure needle irrigation.[13] The vapor lock management also has been shown to remove considerably more debris than manual dynamic irrigation technique from root canals of mandibular molar teeth.[14] The effectiveness of ultrasonic activation of irrigant in cleaning root canals has been confirmed in various studies as well.[15-17]

However, in clinical situation, none of those explanatory variables acts alone, because the cleaning effect of certain irrigation regimen is a combined result of potential influencing factors. To identify the real efficacy of irrigation method systematically, the irrigation technique or device needs to be re-assessed strictly based on the variables affecting the working mechanism in controlled circumstances, not on ambiguous prediction. Therefore, the present study compared the cleaning efficacy of 4 different final additional irrigation regimens in curved canal and isthmus of mandibular posterior teeth. The influences of related variables (continuous replenishment of irrigant, vapor lock management and ultrasonic activation of irrigant) on the cleaning efficacy of the experimented irrigation regimens were also evaluated.

Materials and Methods

Caries free extracted human mandibular molars with fully formed apices were stored in 0.5% chloramine-T solution at room temperature. Mesiodistal and buccolingual radiographs of the teeth were reviewed to evaluate canal curvature according to the Schneider's method, and teeth with 15-45 degrees root canal curvatures were selected for the experiment.[18] Teeth over 45 degrees or multidirectional curvatures were excluded from the study. After examining the canal curvature, the mesial root apex area was carefully examined under the microscope (OPMI Pico, Carl Zeiss Surgical GmbH, Oberkochen, Germany) to select the teeth with two separate apical foramina (mesiobuccal and mesiolingual). Finally selected sixty mandibular molars were accessed with #4 round bur and the access cavities were refined with Endo Z bur. After coronal flaring with #2 to 4 Gates Glidden burs, #10 K file was introduced into the canals as a patency file until it was just visible at the apical foramen. The working length was determined as subtracting 1 mm from the length of the patency files. After working length determination, the apex was sealed with glue and embedded to silicone impression material. Root canal preparation was performed using crown-down technique with Profile .06 instruments (Dentsply Maillefer, Ballaigues, Switzerland) to #35 apical size. Five milliliter of 5.25% NaOCl solution per canal was delivered by 30 gauge Max-i-Probe needle (Dentsply Rinn, Elgin, IL, USA), inserted as far into the prepared root canals as possible without binding during entire instrumentation procedure. Then, the prepared teeth were randomly divided into four experimental groups (n = 15) according to the final irrigation technique used.

Group C (Needle irrigation): Root canals were irrigated with 5.25% NaOCl solution using a 30 G Max-i-Probe for 1 minute per canal. The rate of irrigant supply from the syringe was 6 mL/min. This group was used as a control.

Group U (Ultrasonic irrigation): Root canals were irrigated with aliquots of NaOCl activated by ultrasonics (Suprasson P max ultrasonic device; Satelec, Acteon group, Mérignac, France) for 1 minute per canal (setting: E mode, 25% power). A #15 ultrasonic file was introduced to working length, avoiding contact with canal wall for passive agitation of irrigant.

Group SC (VPro StreamClean irrigation): 30 G side-vent Ni-Ti ultrasonic irrigation tip was activated by ultrasonics (Suprasson P max ultrasonic device; Satelec, Acteon group, Mérignac, France) for 1 minute per canal (setting: E mode, 25% power). The rate of irrigant supply from the syringe was 6 mL/min.

Group EV (EndoVac irrigation): Micro-cannula was passively introduced into each canal to full working length under apical negative pressure for 1 minute per canal according to manufacturer's instruction. The rate of irrigant supply from the syringe was 6 mL/min.

After additional flush, root canals were rinsed with sterile saline and dried with paper points. Mesial roots were separated and immersed in 10% formalin as a fixative for any debris remaining within the canal and isthmus area. Specimens were decalcified and embedded in paraffin block to prepare 4 µm section slides at 1, 3 and 5 mm levels from the apex. The sections were made with Leica RM 2145 microtome (Leica Microsystems, Nussloch, Germany). H&E stained cross-section images were acquired on an Olympus IX-70 microscope (Olympus Corporation, Tokyo, Japan) at 40x magnification using a digital camera (SPOT RT-KE, Spot imaging solutions, Sterling Heights, MI, USA, Figure 1). Spot imaging software (version 4.6, Spot imaging solutions) was used to measure the total area of the canal, isthmus and remaining debris within the spaces. The area of the canal space was calculated (μm^2) by tracing the outline of the root canal. By designating dots in the remaining debris, the area with same color level was

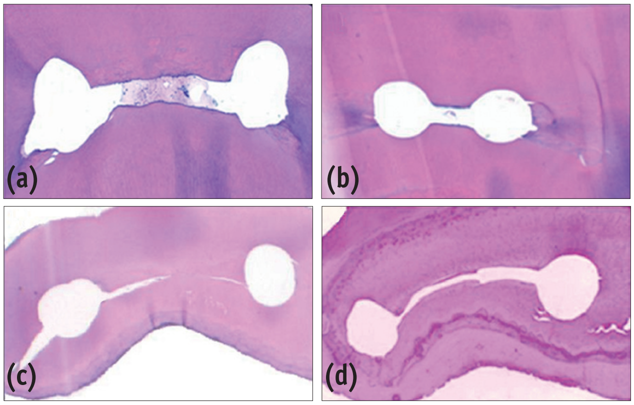

Figure 1. Representative photomicrographs (x40, H&E stain) demonstrating remaining debris at 1 mm level of canal and isthmus in (a) Group C, (b) Group U, (c) Group SC, and (d) Group EV.

automatically captured. By repeating this process, the total debris in the canal space was identified and calculated. Canal cleanliness was determined by subtracting the total area of debris from the canal space area, and the final cleanliness value was expressed in percentage. The isthmus between the canals was traced separately from the primary root canals, and the cleanliness value for the isthmus was also calculated as described above.

The differences of the cleanliness values among groups were compared using Kruskal-Wallis test and Mann-Whitney U test. To identify a hypothetical model for cleaning efficacy of irrigation technique, experimental groups were re-categorized according to the contributing variable as follows: continuous replenishment of irrigant, vapor lock management, and ultrasonic activation of irrigant (Table 1). Multiple linear regression analysis was used to identify a hypothetical model and thus to find significant variables affecting the cleaning efficacy at each level of isthmus. All the analyses were performed using SPSS 17.0 (SPSS Inc., Chicago, IL, USA), and the significance level was set at α = 0.05.

Results

All four groups demonstrated higher cleanliness values in root canals than those in isthmi (Table 2). The mean cleanliness values of the canal and isthmus according to the irrigation technique at each level are shown in Figure 2.

Table 1. Re-categorizing of the experimental groups according to the variables. The presence or absence of the variables was marked as “1” or “0”, respectively

	Group C	Group U	Group SC	Group EV
Regular replenishment	1	0	1	1
Vapor lock management	0	0	0	1
Ultrasonic activation	0	1	1	0

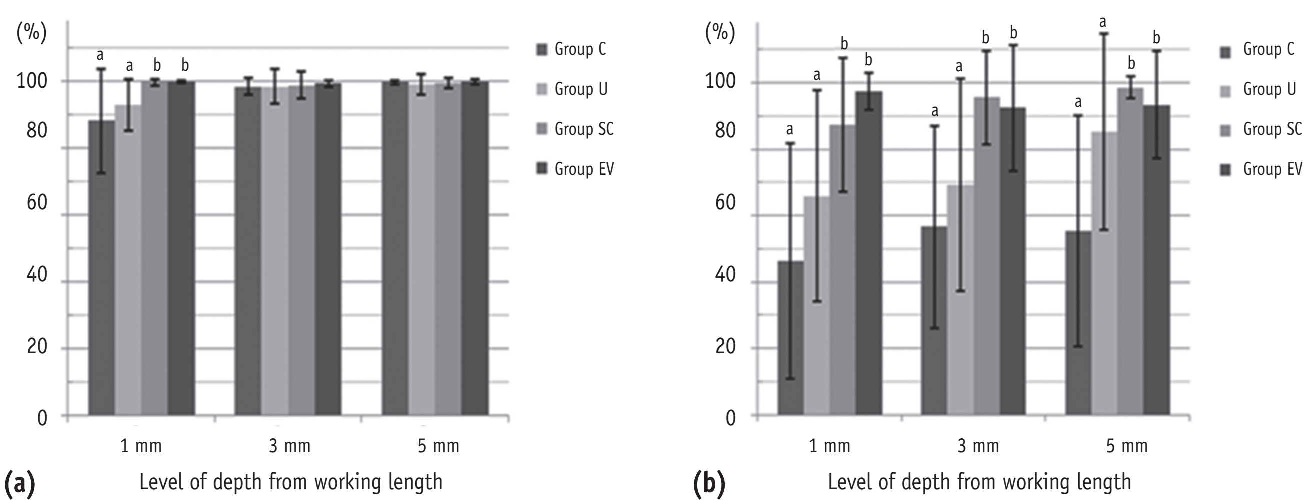

Figure 2. Comparison of cleanliness values among experimental groups at each level of the depth. (a) Canal cleanliness values; (b) Isthmus cleanliness values.
* Statistically significant differences are shown by superscript letters. Same letters are not significantly different ($p < 0.05$).

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Table 2. Overall cleanliness values of canal and isthmus

	Group C	Group U	Group SC	Group EV
Canal cleanliness (%)	95.40 ± 9.89	99.20 ± 6.20	99.38 ± 2.55	99.70 ± 0.78
Isthmus cleanliness (%)	69.46 ± 31.87	73.50 ± 31.66	93.90 ± 13.96	94.45 ± 14.71

Table 3. Result of multivariate linear regression analysis of canal cleanliness

	R	Coefficients (*p* value)			
		Regular replenishment	Vapor lock management	Ultrasonic activation	Constant
1 mm	0.516	6.797 (0.030)	11.782 (0.000)	11.623 (0.001)	81.334
3 mm	0.114	0.307 (0.812)	0.927 (0.474)	0.250 (0.847)	62.318
5 mm	0.128	0.347 (0.596)	0.085 (0.896)	0.150 (0.819)	99.343

Table 4. Result of multivariate linear regression analysis of isthmus cleanliness

	R	Coefficients (*p* value)			
		Regular replenishment	Vapor lock management	Ultrasonic activation	Constant
1 mm	0.630	31.653 (0.041)	41.075 (0.001)	51.228 (0.000)	24.775
3 mm	0.568	26.420 (0.011)	35.829 (0.001)	39.065 (0.000)	30.217
5 mm	0.566	13.273 (0.135)	37.934 (0.000)	43.188 (0.000)	42.172

The cleanliness values of both canal and isthmus at 1 mm level were significantly lower than those of 3 and 5 mm level. Comparing canal cleanliness, group SC and group EV showed significantly higher cleanliness values than group C and group U at 1 mm from the apex ($p < 0.05$). When isthmus cleanliness was evaluated, group SC and group EV demonstrated significantly higher cleanliness values than those of group U at 3 mm level and group C at all levels ($p < 0.05$). The isthmus cleanliness value of Group U was higher than that of group C at only 5 mm level ($p < 0.05$).

The results of the multiple linear regression analysis are shown in Table 3 and 4. The variables showed independent positive correlation with canal cleanliness at 1 mm level. The vapor lock management was the most influencing factor for the debridement of canal at 1 mm level, followed by ultrasonic activation and continuous replenishment of irrigant. On the other hand, 3 variables had independent positive correlations with isthmus cleanliness at all levels. Of the 3 variables, the order of influence on isthmus cleanliness was ultrasonic activation of irrigant, vapor lock management, and continuous replenishment of irrigant in descending order. The multiple correlation coefficient (R value) was greatest at 1 mm level of isthmus (R = 0.630), and the ultrasonic activation of irrigant had greater coefficient than the other variables at this level.

Discussion

A number of studies have investigated the effectiveness of various irrigation regimens. Ultrasonics activates the flow of irrigant by acoustic microstreaming and cavitation, and its effectiveness in cleaning root canal has been proven in various studies.[15,16,19-21] VPro StreamClean is expected to enhance removing debris from root canal system by activating the refreshing irrigants continuously with ultrasonic power. Major problem of these two irrigation regimens is that it is impossible to ensure whether the irrigant reaches to full working length. In this respect, apical negative pressure which is applied in EndoVac system enables eliminating vapor lock area and pulls fresh new irrigant into the apical region. This promotes effective flushing action in terms of safety and irrigant volume. However, there has been no in depth verification of these irrigation regimens with their own hypothetical variables. The re-assessment of irrigation techniques or devices based on related variables affecting the working mechanism may propose the most meaningful explanation for selecting proper irrigation regimen at certain clinical situation. Therefore, the real effectiveness of specific irrigation regimen in relation to hypothetical variables; vapor lock management, ultrasonic activation of irrigant, and continuous replenishment of irrigant; is the subject of interest in this study.

All groups showed over 98% mean canal cleanliness values at 3 and 5 mm level from the apex, and there were no statistically significant differences among the groups. This result is in consistent with earlier studies which reported the "needle deep" irrigation, inserted as deep as possible to working length, is important in improving canal cleanliness.[21,22] Moreover, there was no difference in canal cleanliness between ultrasonic irrigation and conventional needle syringe irrigation. This finding is in contrast to previous studies in that the ultrasonically activated file was effective in debris removal from all levels of the root canal.[15,19,23] It is perceived that this ineffectiveness of ultrasonic irrigation is partly related to the apical vapor lock phenomenon. Once an ultrasonically activated tip leaves the irrigant and enters the vapor locking area at apical 1 mm, acoustic microstreaming and/or cavitation becomes physically impossible. Smaller canal diameter below the curvature might be another reason for the limited effect of ultrasonic irrigation.

The differences among experimental groups were more remarkable in isthmus, where the debridement might be solely accomplished by flushing action of irrigants. In this study, VPro StreamClean and EndoVac system showed higher cleanliness values at all levels of isthmus while ultrasonic irrigation showed significantly higher values only at 5 mm level of isthmus compared to that of needle-syringe irrigation. Regarding the correlation coefficients of the hypothetical variables of the experimental groups, continuous delivery of fresh irrigant was a relevant variable to produce higher cleanliness values especially in the isthmus rather than the root canals. Considering the consuming time of chlorine from NaOCl solution, the replenishment of sufficient amount of irrigant produced by VPro StreamClean and EndoVac system accounts for their higher cleaning efficacy.[24]

Multi-variable statistical approach provided overall contribution of each independent variable on cleaning efficacy despite of the complexity of the influencing variables. In canal cleanliness, the variables had significant correlation at 1 mm level, as in the result of comparison among groups. The constant was relatively higher than that of isthmus, which indicates relatively lower differences among groups. On the other hand, the influence of the variables was significant at all levels of isthmus, and the ultrasonic activation of irrigant was the most impacting factor among them despite subtle differences of coefficients according to the depth. This hypothesized model to predict the cleaning efficacy of specific irrigation technique may not fit in all clinical cases due to the variability of width or length of isthmus. However, considering R values of the model, the remarkable differences among groups at 1 mm level could be explained.

Although the result of this study seems to imply predictable debridement capacity of VPro StreamClean and EndoVac system, the limitation of the study design should not be overlooked; efficacy of irrigation measured as percentage of cleaned cross-sectional area in the canal lumen does not imply the bacteria-free environment or cleaned dentinal tubules on root canal walls. Peters *et al.* reported that instrumentation techniques using rotary Ni-Ti files left 35% or more of the canals' surface area of molar teeth unchanged.[25] Therefore, the additional use of root canal irrigation and intracanal medicament in infected root canals having isthmus is strongly suggested for a suitable period of time.[26,27] Further investigations on the possible differences of isthmus cleanliness within the experimental groups caused by the variations in isthmus width and length are required as well.

Conclusions

In conclusion, apical vapor lock management is the most important factor in canal cleanliness. If inter-canal communications such as isthmus is present, ultrasonic activation of irrigant is the first technique of choice. Combined application of vapor lock management and ultrasonic activation might be advantageous as an additional irrigation method. In this regard, both VPro StreamClean and EndoVac system can be effective irrigation regimens for cleaning debris in the complicated root canal system having curved canal and/or isthmus.

Conflict of Interest: No potential conflict of interest relevant to this article was reported.

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