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EFFECT OF DIFFERENT IRRIGATION SYSTEMS ON SEALER PENETRATION INTO DENTINAL TUBULES

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Keywords

Confocal laser scanning microscopy; dentinal tubules; irrigation; sealer penetration; Thermafil Obturator

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Abstract

Introduction: Different irrigation systems have been developed to improve the efficacy and distribution of the irrigants. The aim of this study was to compare the effect of conventional endodontic needle irrigation, with other irrigant delivery and/or agitation systems on sealer penetration into dentinal tubules.

Methods: Fifty single-rooted teeth with round-shaped root canals were distributed in 5 homogeneous groups characterized by the different cleansing system used: conventional endodontic needle irrigation, EndoActivator, Irrisafe, Self-Adjusting File and EndoVac. After instrumentation, all teeth were filled by Thermafil Obturators and Rhodamin B dye labelled TopSeal sealer. Teeth were transversally sectioned at 2-, 5-, and 7-mm levels from the apex and observed under confocal laser scanning microscope. Maximum, mean and percentage of sealer penetration inside tubules around the root canal were measured. Moreover, the integrity of the sealer layer perimeter was evaluated.

Results: No significant differences both in mean ($P > 0.05$) and in maximum penetration depth ($P > 0.05$) were observed among groups, while both parameters showed an increased trend within each group from the 2- to the 7-mm level from apex. Similarly, the percentage of penetration around the root canal wall did not differ among groups ($P > 0.05$) and showed an increasing trend within each group from the apical to the coronal portion of the canal.

Conclusions: Sealer penetration into dentinal tubules is not affected by the irrigant delivery and/or agitation systems studied. Thermafil with TopSeal technique achieves the complete sealer perimeter integrity in all groups.

Introduction

Needle irrigation is the conventional method to deliver irrigants inside the root canal system (RCS), but to reach the full length of the root canal, the needle tip has to be inserted within 1-mm of working length (1) increasing the risk of irrigants extrusion from apical foramen. Nonetheless, the vapor lock phenomenon might prevent the direct contact of the irrigant with the root canal wall, especially in its most apical portion, thus making the irrigant action ineffective (2). Therefore, to improve the efficacy and distribution of the irrigants different irrigation techniques and devices have been developed, such as EndoActivator, Irrisafe, Self-Adjusting File and Endovac. EndoActivator (Dentsply, Tulsa Dental Specialities, Tulsa, OK) is a sonic device that employs frequencies in the ranges of 2-3 kHz to activate irrigant solutions. It has been reported that this device produces a hydrodynamic activation of the irrigants that is able to safely clean the RCS and morphological irregularities as lateral canals and apical deltas (3). Irrisafe (Satelec Acteon Group, Merignac, France) is an ultrasonic device operating in the range of 25-30 kHz that activates the irrigant solution by acoustic streaming and micro-cavitation; this technique is referred as passive ultrasonic irrigation (PUI) and it allows the delivery of irrigants up to the working length of the root canal unlike conventional endodontic needle (4). The Self-Adjusting File (ReDent Nova, Ra'anana, Israel) is a hollow and flexible file that adapts itself three-dimensionally to the root canal. During the instrumentation technique, this file allows for the continuous irrigation that, in combination with the vibrating motion, influences cleaning ability in the root canal, particularly in the apical third (5). The EndoVac (KerrEndo, Orange County, CA) is an apical negative pressure irrigation system that sucks the irrigant solution by means of a microcannula positioned at the working length. Thus, the vapor lock effect and the risk of NaOCl extrusion beyond the apical foramen are prevented. Moreover, the EndoVac has been shown to improve the cleaning of the apical third with respect to conventional needle irrigation (6).

The aim of the present study was to compare the effect of conventional endodontic needle irrigation and four different irrigation systems on sealer penetration into dentinal tubules of extracted teeth with round root canal using confocal laser scanning microscopy (CLSM). The null hypothesis tested was that there is no difference in the depth and percentage of sealer penetration between the conventional endodontic needle irrigation, and four different method of root canal cleaning. Moreover, the integrity of the sealer layer perimeter was evaluated.

Materials and Methods

Fifty human teeth with single round-shaped root canals and straight mature roots were selected from a pool of extracted teeth.

After access cavity preparation, the working length (WL) was established by subtracting 1 mm from the total root length. The apex was covered with cyanoacrylate to simulate in vivo conditions. Samples were distributed into 5 experimental groups of 10 teeth each and characterized by the different systems used: conventional endodontic needle irrigation group (CENI); EndoActivator group (EA); Irrisafe group (IS); Self-adjusting File group (SAF) and EndoVac group (EV). Canal width, measured on radiographs at 5 mm from the apex, and WL were not different among groups (Kruskal-Wallis test, $P > 0.05$). Each canal was instrumented in a crown-down manner using the ProTaper Universal rotary system (Dentsply Maillefer, Ballaigues, Switzerland) to size 40 at the WL.

In the CENI group, after the use of each instruments, the root canals were irrigated with 1 mL 5.25% NaOCl (Niclor, Ogna, Muggiò, Italy) using a syringe with a 30-G side-vented needle (Max-i-Probe, Dentsply Rinn, Elgin, IL) placed before the binding point but not closer than 2 mm from the WL. After instrumentation, the canals were finally rinsed with 1 mL 5.25% NaOCl left in place for 30 seconds followed by 1 mL

17% EDTA (Ogna, Muggiò, Italy), left in place for 30 seconds and with 1 mL 5.25% NaOCl left in place for another 30 seconds. The needle was placed 2 mm from WL.

In the EA group, the irrigation protocol was the same as the CENI group but the final irrigation was performed using the 25 .04 non cutting polymer tip of the EndoActivator, placed 2 mm from the WL for 30 seconds for each irrigant solution.

In the IS group, the irrigation protocol was the same as the CENI group but the final irrigation was performed using a stainless steel non cutting 25 tip (Irrisafe, Satelec, Acteongroup, Merignac, France) mounted on an ultrasonic device (P5 Newtron, Satelec), placed 2 mm from the WL for 30 seconds for each irrigant solution.

In the SAF group, the irrigation protocol was the same as the CENI group but the final irrigation was performed using the 1.5-mm SAF file (ReDent-Nova, Ra'anana, Israel). The SAF file was operated by using an in-and-out manual motion for 30 seconds (0.4-mm amplitude and 5000 vibrations per minute) with continuous irrigation by using 5.25% NaOCl provided by a VATEA peristaltic pump (ReDent-Nova) at a rate of 2 mL/min. A second cycle was performed as just described but employing 17% EDTA, and the third and last cycle with 5.25% NaOCl.

In the EV group, after each instrument change, 1 mL of NaOCl was delivered to fill the access cavity. At the end of instrumentation, NaOCl 5.25% was delivered with the macrocannula for 30 seconds with an up and down movement from a point where it started to bind to a point just below the canal orifice. NaOCl was left in place for 60 s then 3 cycles of microirrigation were performed inserting the microcannula at WL for 6 s, then at 2 mm from WL for 6 s, and eventually at WL for another 6 s. This was done until a total of 30 s was reached, for each cycle. At the end of cycles, the microcannula completely aspired the irrigant from within the canal. The first and third cycles were performed using NaOCl 5.25% while the second cycle employed 17% EDTA. In all groups the same amounts of irrigants were employed.

All canals were dried with paper points and filled by Thermafil Obturators 40 (Dentsply, Tulsa Dental Specialities, Johnson City, TN, USA) with TopSeal sealers (Dentsply De Trey, Konstanz, Germany) labeled with 0.1% wt Rhodamin B dye

(Carlo Erba Reagenti, Arese, Italy). The sealer was introduced into the canal by means of a paper point 40 to 1 mm short of the WL in a pumping motion for 5 s. A coronal filling was performed with a temporary material (Cavit, 3M ESPE, Seefeld, Germany) and then teeth were stored in an incubator at 37°C and 100% humidity for 7 days to allow the sealer to set.

The teeth were embedded in methacrylate resin (Technovit 3040, Heraeus Kulzer, Wehrheim, Germany) and transversally sectioned at 2-, 5- and 7-mm from the apex with a saw microtome (Leica SP 1600, Nussloch, Germany) to obtain 200-μm-thick sections. These were examined under CLSM (Leica TCS SP2 AOBS, Mannheim, Germany) at 5 X and 10 X magnification. The depth of sealer penetration into dentinal tubules was calculated as the average penetration measured, using the straight-line tool of ImageJ software (National Institutes of Health, USA), at 8 standardized points starting from the inner side of canal wall at 2-, 5- and 7-mm from the apex (7). Moreover, the point of deepest penetration was measured from the canal wall to the point of maximum depth of sealer penetration. The percentage of sealer penetration was calculated by measuring the Rhodamine B stained surfaces of the canal wall where sealer penetrated inside dentinal tubules (sealer tags) and dividing these values by the circumference of the root canal itself and multiplying the result by 100. Moreover, the integrity of the sealer layer perimeter was evaluated on each image acquired measuring the Rhodamine stained perimeter of the canal wall and dividing this value for the root canal circumference and expressed as percentage.

Comparisons among groups in sealer penetration (expressed as mean, maximum and percentage penetration) were performed using the nonparametric Kruskal-Wallis test, allowing post hoc pairwise multiple comparisons when appropriate. Differences within each group in sealer penetration at 2-, 5- and 7-mm levels were analyzed by using the Wilcoxon signed rank sum test. Statistical analyses were performed using IBM SPSS Statistics package ver. 21 and a P values < 0.05 was considered significant.

Results

Figure 1 shows an overview of representative CLSM images from each experimental groups at 2-, 5- and 7-mm levels.

Sealer penetration (Figure 2) was not different among groups (Kruskal-Wallis test, $P > 0.05$), while, within each group, mean depth of sealer penetration showed an increasing trend from the apical toward the coronal third. More specifically, within each experimental group, at the 2-mm level the results were significantly lower compared to those measured at 5- and 7-mm level (Wilcoxon signed rank sum tests, $P < 0.05$). Further, in the SAF group, the mean depth measured at 5-mm level appeared significantly lower than that measured at 7-mm level (Wilcoxon signed rank sum test, $P < 0.05$).

Table 1 reports the average values of maximum depth of sealer penetration (μm) recorded at each level for each group. No statistically significant differences were observed among groups (Kruskal-Wallis test, $P > 0.05$), while values measured at 2-mm levels were always smaller compared to those observed at 5- and 7-mm levels (Wilcoxon signed rank sum tests, $P < 0.05$), within each group, except for EA group.

In this group values at 2-mm were not significantly lower than those measured at 5-mm level (Wilcoxon signed rank sum tests, $P > 0.05$), while values observed at 5-mm were significantly lower than those measured at 7-mm level (Wilcoxon signed rank sum test, $P < 0.05$).

In SAF group, on the contrary; differences in maximum depth observed among levels were always statistically significant (Wilcoxon signed rank sum tests, $P < 0.05$).

The percentage of sealer penetration into dentinal tubules (Fig. 3) was not significantly different (Kruskal-Wallis test, $P > 0.05$) among groups when the overall distribution of values was compared. Within each group, an increase in the percentage of sealer penetration was observed from the apex toward the coronal third. Percentages of penetration measured at 2-mm level appeared always significantly lower than those recorded at 5- and 7-mm levels (Wilcoxon signed rank sum tests, P

< 0.05), and, both in EndoVac and in SAF group, percentages observed at 5-mm level resulted significantly lower than those measured at 7-mm level (Wilcoxon signed rank sum tests, $P < 0.05$), while in remaining groups no significantly differences could be measured between 5- and 7-mm levels (Wilcoxon signed rank sum tests, $P > 0.05$).

The integrity of the sealer layer perimeter was 100% in all groups and in all levels examined.

Discussion

Sealer penetration into dentinal tubules is considered a positive outcome in order to: prevent bacterial repopulation or bacterial inactivation inside the tubules as a blocking agent (8), improve filling material retention within the root canal thanks to mechanical interlocking between sealer and root dentin (9), entomb remaining bacteria within dentinal tubules (10). Therefore, sealer penetration into dentinal tubules is considered clinically relevant (11).

No studies have been performed so far about sealer penetration into dentinal tubules comparing, at the same time, different activation/delivery irrigation systems in association with a sealer and a core-carried based technique (Thermafil) by using CLSM analysis. Few studies (7,12,13), based on CLSM analysis, evaluated sealer penetration into dentinal tubules using the Thermafil technique. Only two studies using Thermafil (12,13) employed an activation of the irrigants by means of passive ultrasonic irrigation.

In this study no significant differences were found in all investigated parameters related to sealer penetration in association with the different systems evaluated. These results are similar to those reported by Bolles (14), who showed that sonic activation of irrigants by means of EndoActivator and Vibringe did not significantly improve sealer penetration with respect to conventional irrigation. The EndoActivator system does not increase smear layer removal compared to conventional irrigation (15).

Contrary to our results, Kara Tuncer & Unal (16) show that the EndoVac irrigation system improves sealer penetration with respect to conventional irrigation at 1- and 3-mm levels from the apex. Sealer penetration cannot be reputed an absolute index of smear layer removal, because the presence of smear layer limits, but do not completely prevents sealer penetration into tubules (11). Therefore, sealer penetration into dentinal tubules might more likely depend on its physico-chemical properties (17) and on the filling technique employed rather than on the activation of irrigants. In this study, TopSeal, an epoxy resin-based sealer, has been employed with a core-carried filling technique. AH Plus (TopSeal) sealer has a pseudoplastic behavior characterized by a reduction of viscosity and an increase in flow when shear rate increases during the filling procedure (18). This physical property allows the sealer to adhere to the root canal wall, to fill uninstrumented accessory root-canals and to penetrate dentinal tubules. In case of retreatment, an epoxy resin based sealer is impossible to remove completely from dentinal tubules, because it deeply penetrates into dentinal tubules and its removal would imply a removal up to 60 % of additional root dentin. Nonetheless, the complete removal of the sealer from dentinal tubules does not mean a retreatment failure (19).

In all our sections examined, the integrity of the sealer layer perimeter was 100%, thus indicating an optimal adaptation of the sealer to the canal wall. This is probably due, not only to the physico-chemical characteristics of the sealer, but also to the core-carried filling technique employed. Also Kok (12) found that the integrity of the sealer layer was 100% when AH Plus sealer was associated with the Thermafil technique, while it was 98.43% with the cold lateral compaction technique and 99.54% with the single master cone one. Core-carried techniques, in fact, allow a homogeneous root filling where the sealer layer is thin and well adapted to the canal wall (20).

In this study all investigated parameters related to sealer penetration show an increasing trend from the apical to the coronal level. Our results are in line with those reported by recent investigations (7, 21-23). The minor penetration here observed at the apical level might be ascribed to the reduction of tubular diameter and number towards the apex, and to dentinal sclerosis (24).

Conclusion

The use of either EndoActivator or Irrisafe or Self-Adjusting file or Endovac systems in round-shaped straight root canals does not improve sealer penetration into dentinal tubules with respect to conventional endodontic needle irrigation when TopSeal sealer is associated with the Thermafil Obturator technique.

This filling method ensured an integrity of the sealer layer perimeter of 100% independently of the device employed.

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Figure legends

Figure 1 Representative CLSM images from each experimental groups at 2-, 5- and 7-mm levels.

Figure 2 Mean depth of sealer penetration (μm) measured at different levels within each experimental group. In each boxplot, the median value (the line inside the box), the interquartile range (IQR length of the box), the minimum and maximum values (extreme lines) are reported. Values more than 1.5 IQR's but < 3 IQR's are labeled as outliers (o); values ≥ 3 IQR's are labeled as extreme cases (*).

Figure 3 Percentage of sealer penetration measured at different levels within each experimental group. In each boxplot, the median value (the line inside the box), the interquartile range (IQR length of the box), the minimum and maximum values (extreme lines) are reported. Values more than 1.5 IQR's but < 3 IQR's are labeled as outliers (o); values ≥ 3 IQR's are labeled as extreme cases (*).

Figure 1

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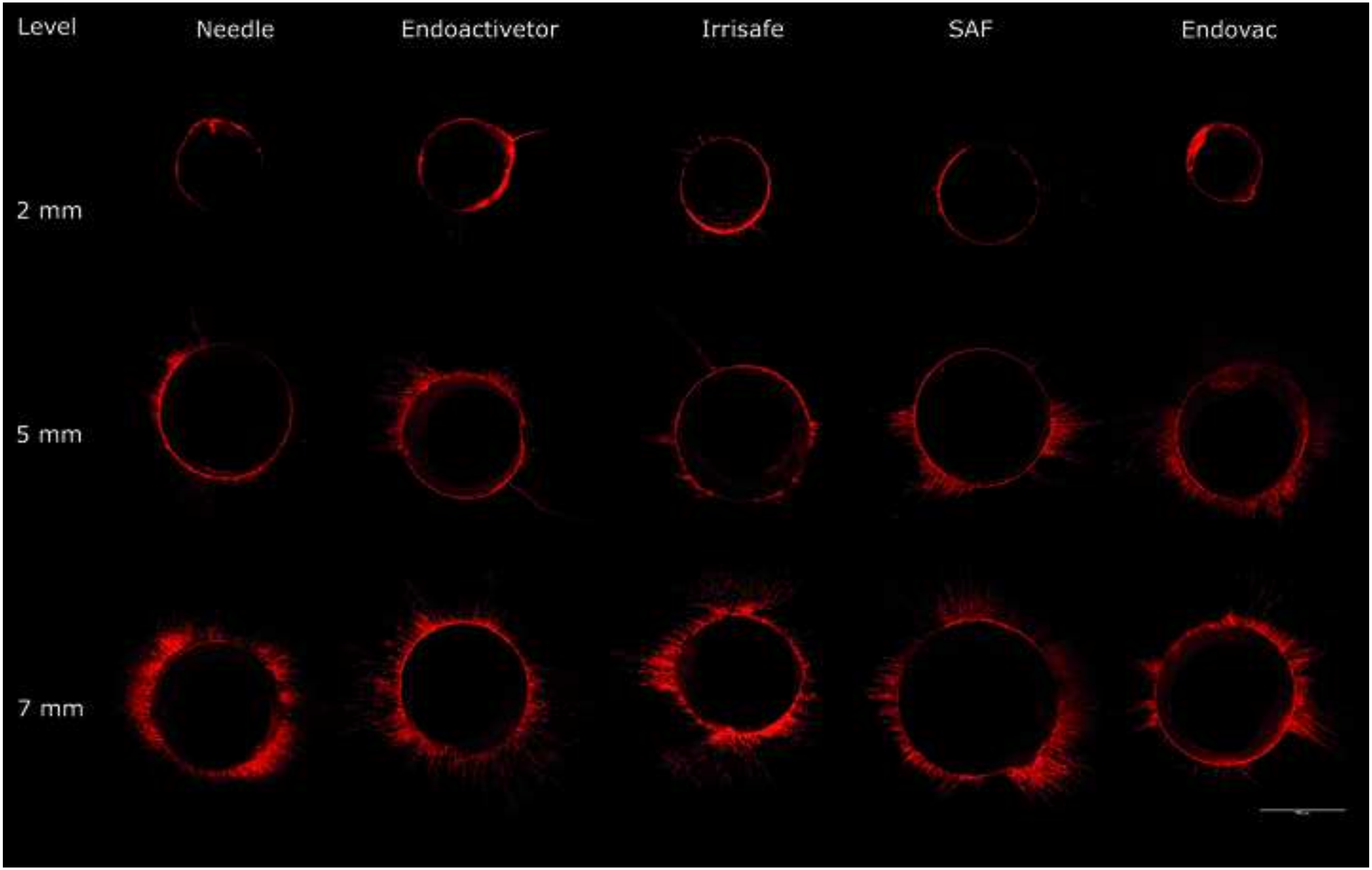

Figure 2
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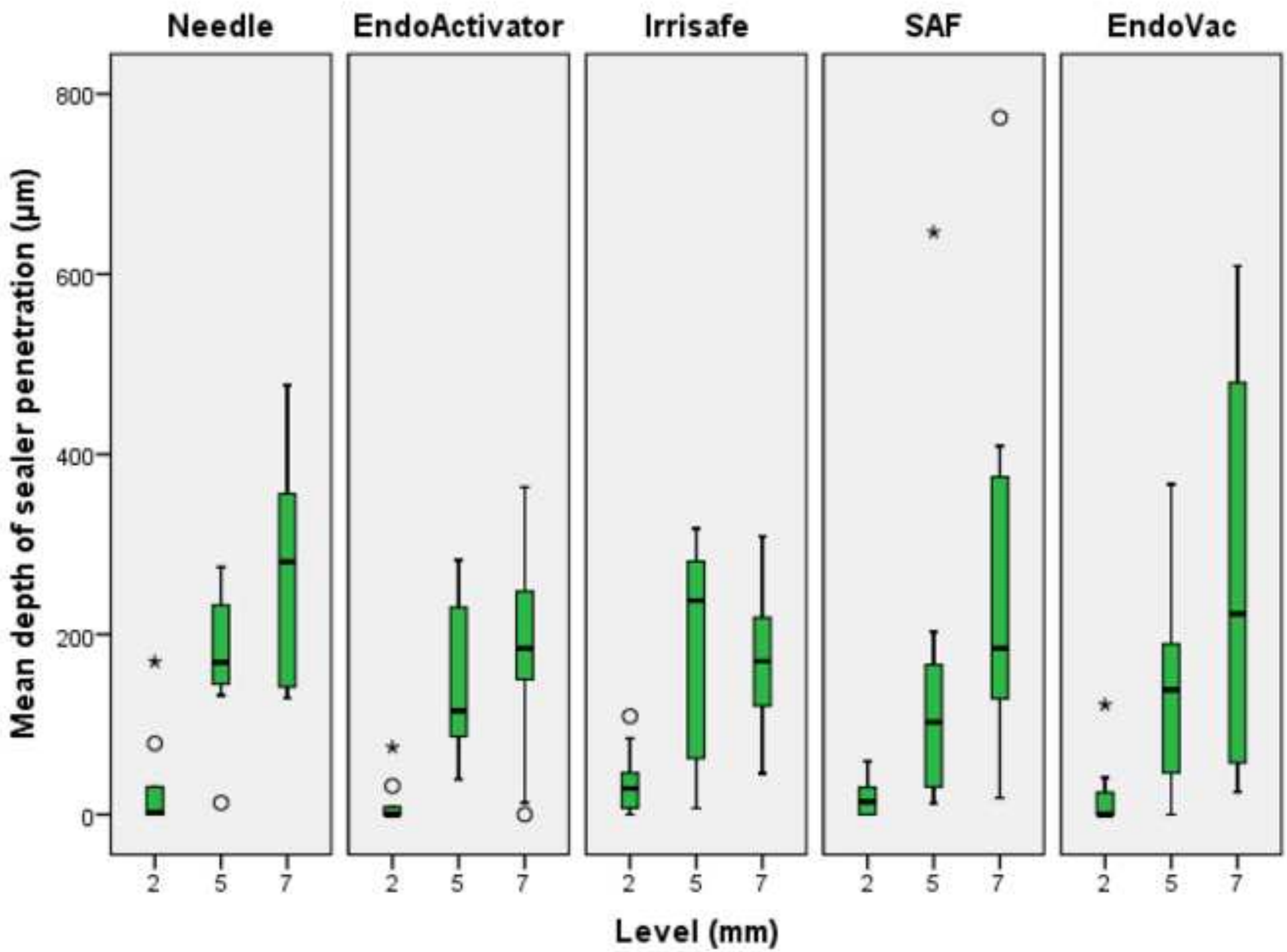

Table 1 Maximum depth of sealer penetration (mean ± SD, µm) recorded at each level with each experimental group

group	level		
	2-mm	5-mm	7-mm
CENI	288 ± 403 [a,b]	987 ± 374[a]	1012 ± 269[b]
EA	348 ± 402 [b]	658 ± 298[c]	900 ± 352 [b,c]
IS	401 ± 205 [a,b]	756 ± 420 [a]	844 ± 228 [b]
SAF	215 ± 230 [a,b]	570 ± 469 [a,c]	858 ± 498 [b,c]
EV	214 ± 252 [a,b]	703 ± 466[a]	963 ± 324[b]

[a] 2-mm vs 5-mm level: Wilcoxon test $P < 0.05$; [b] 2-mm vs 7-mm level: Wilcoxon test $P < 0.05$; c 5-mm vs 7-mm level: Wilcoxon test $P < 0.05$.

Figure 3

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