Coronal Leakage of two Different Root Canal Sealers

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Abstract
The objective of this study was the evaluation and comparison of coronal bacterial leakage of two different instrumentation techniques and two endodontic sealers Apex Plus and EndoRez.

Sixty-eight human teeth caries free were tested in this study. Cleaning and shaping was carried out with step-back and crown-down technique and irrigated with 2% NaOCl and 17% EDTA. After instrumentation roots were randomly divided into four groups, two control groups (positive and negative) and two experimental groups according to root canal sealer: thirty-two teeth of first group were obturated with Apex Plus and thirty-two teeth of second group were obturated with EndoRez. These groups (1st and 2nd) were further subdivided into two subgroups (a and b) according to the root canal instrumentation technique. After setting time, the roots were incorporated in a leakage model a two chamber method and research on microleakage was conducted for 33 days.

The minimum microleakage values were obtained from the teeth obturated with EndoRez sealer and instrumented with crown-down technique. There were significant differences between the subgroups instrumented with step-back technique in terms of microleakage (T-testi=4.734, p=0.009).

Neither instrumentation technique nor endodontic sealers could prevent coronal microleakage.

Keywords: Leakage, instrumentation technique, Apexit Plus, EndoRez.

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Introduction
Successful endodontic treatment is achieved by the proper cleaning and shaping of the root canal, and the complete and hermetic obturation of the root canal system. Manual and rotary instrumentation is one of the important contributors to remove all necrotic and vital organic tissue and give the canal system a shape that facilitates placement of permanent root filling. Obturation of the root canal is an essential part of endodontic treatment and must be performed to the highest clinical standards.

Sealed root canals can be recontaminated under several circumstances: (a) if the patient has had delayed placement of permanent restorations; (b) if the seal of the temporary filling material has broken down; or (c) if filling materials and/or tooth structures have fractured. The sealing properties of root canal filling materials are another important factor that can influence the success of treatment. Root canal obturation materials in order to prevent leakage requires certain clinically conditions, including quality cleaning and shaping the canal system, most of them requires perfectly dry canals to achieve good adhesion, skills and experience of the clinician and health condition of the supporting periodontium. The most widely used root canal filling material is gutta-percha. Gutta-percha, since it is a material that lacks elasticity and does not bond to root dentin, must be used in conjunction with sealer cement.

Apexit Plus (Vivadent, Schaan, Liechtenstein) is a calcium hydroxide type sealer, which may have good sealing ability and biocompatibility with tissues. Apexit Plus contains calcium hydroxide, calcium phosphate, zinc oxide, silicon oxide, plasticizer, disalicylate esters, and bismuth. The aim of using calcium hydroxide as a root canal sealer is stimulation of the periapical tissues in order to maintain health or promote healing and secondly for its antimicrobial effects. Ozata et al. (1999)
compared the apical leakage of Ketac-Endo, Apexit and Diaket and found that there was no significant difference between Apexit and Diaket. Using coronal bacterial leakage, Timpowat et al. (2001), concluded that canals filled with a Ca(OH)2-based sealer was significantly greater than those filled with a resin-based sealer.

EndoRez (Ultradent Inc, South Jordan,UT) is a hydrophilic, urethane-dimethacrylate–based resin sealer. Recent studies have shown that EndoREZ, provides an effective seal when used with lateral condensation. The hydrophilic properties of the sealer allow penetration deep into accessory canals and dentinal tubules but not into gutta-percha. According to Drukleinis et al. and Ulusoy et al., based on their study they concluded that root canal samples filled with EndoRez have shown minimum leakage.

The purpose of this study was to compare the coronal leakage of two instrumentation technique and two root canal sealers, using Enterococcus faecalis to penetrate through the obturated root canal. The teeth were observed for bacterial leakage on daily basis for thirty-three days.

Materials and methods

Specimen Selection
Sixty-eight caries free, human maxillary incisors and canines with straight roots were used in this study. All teeth were stored in a sealed container with the sterile saline solution prior to the study.

Instrumentation of root canals
Working length was determined by placing size 10 K-file (Maillefer, Ballaigues, Switzerland) into the root canal until its tip was visible at the apical foramen. The root canals were prepared 1mm short of this length. The cervical and middle thirds were prepared with Gates Gliidden drills sizes 2 and 3 (Maillefer, Dentsply, Switzerland). Thirty- four canals were prepared according to a crown-down technique with ProTaper rotary system (Dentsply, Maillefer, Ballaigues, Switzerland), preparation of the middle and apical sectors of the canal up to the working length with S1, S2 shaping files and F1, F2, and F3 finishing files. The X-Smart endodontic motor (Dentsply, Maillefer) was used at a rotation 250 rpm, introducing the instruments passively into the root canal. The other thirty-four canals were prepared with stainless steel K-files (Diadent, France) using a step-back technique. Apical enlargement was made with instrument size up to no. 30.

After each file, the root canal was irrigated with 2% NaOCl (Cerkamed, Poland). Upon completion of instrumentation, the smear layer was removed from the canals by rinsing with 5 ml of a 17-percent EDTA (Cerkamed, Poland) solution for 1 minute, and then the canals were rinsed with 5 ml saline solution to eliminate any residual chemical effects of irrigating solutions, and dried with papers points. To prevent dehydration, all roots were handled using water-moistened gauze during instrumentation. The roots were sterilized by immersing them in Gigasept PA (Schülke UK, Sheffield, UK).

Obturation of root canals
The roots were divided into 2 experimental groups (n=32) according to the sealer: G1-Apexit Plus (Vivadent – Schaan, Liechtenstein), G2-EndoRez (Ultradent – South Jordan, Utah, USA). These groups (1st and 2nd) were further subdivided into two subgroups (a: 16 teeth and b: 16 teeth) according to the canal preparation technique.

Positive and Negative Control Groups
The positive control included two teeth that had a single cone with no sealer (G3). This allowed free communication of the bacteria in the upper chamber with the growth medium in the lower chamber. The negative control consisted of two teeth with one tooth obturated with only Apexit Plus and one tooth obturated with only EndoRez (G4). The samples in this group were coated with a bonding agent to seal the apical opening and the dentinal tubules to prevent any leakage.

Leakage Apparatus Preparation.
A microbial leakage apparatus was constructed using a similar two-chamber method described by Torabinejad et al. (1990). A 20-ml scintillation vial was modified to create a dual-chamber device. Each tooth was placed in the center of each plastic cap and secured using sticky wax. The lower chamber of the apparatus, was filled with sterile tryptic soy broth (TSB) containing streptomycin (2000 µg/ml). The upper chamber of the apparatus, that space above the canal orifice of the tooth, was
filled with Enterococcus faecalis ATC 51299 (Liofilchem, Roseto degli Abruzzi, Italy). Fresh medium and E. faecalis were added to the upper chamber every four days. All of the experimental samples were placed in a 5.0-percent CO2 incubator at 37°C and 100-percent humidity. The microleakage experiment was conducted for 33 days and the medium in the bottom chamber was examined daily for turbidity changes.

**Statistic analysis**

Data obtained from the study was analyzed using Turkey’s HSD test and T-test of proportion. In this study, statistical significance was set at P≤0.05.

**Results**

The positive control group (G3) presented contamination (turbidity) within the first 24 hours, and the negative control group (G4) presented no turbidity throughout the entire experimental period (33 days).

According to cumulative frequency analysis microleakage during the root canal sealing with Apexit Plus in first 5 days, actually in first 12 days, sealing conducted using crown–down technique was higher comparing to step-back technique, but this difference wasn’t statistically significant (12.5% vs 0.0%, p=0.15).

Maximum leakage using step-back technique is achieved at day 26, in two cases (12.5%), whereas the highest scale of turbidity using crown-down technique is achieved already in second day, also in two cases (12.5%). Therefore, maximum scale of coronal leakage from root canal system sealed with Apexit Plus was identical during step-back and crown-down sealing techniques (T-test of proportion =0.0, df=30,p=1.0) (Tab 1 and graph 1).

<table>
<thead>
<tr>
<th></th>
<th>Step-back technique (n=16)</th>
<th>Crown-down technique (n=16)</th>
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<tbody>
<tr>
<td>Day</td>
<td>Cumulative frequency</td>
<td>%</td>
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</tr>
<tr>
<td>1-5</td>
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<td>0.0%</td>
</tr>
<tr>
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<tr>
<td>13-19</td>
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<tr>
<td>20-26</td>
<td>2</td>
<td>12.5%</td>
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<tr>
<td>27-33</td>
<td>2</td>
<td>12.5%</td>
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NT – not tested

**Table 1.** Presentation of coronal microleakage from samples treated with step-back and crown-down techniques and sealed with Apexit Plus.

Microleakage in first five days during canal obturation using EndoRez, was higher at step-back obturation technique, comparing to crown-down, but this difference wasn’t statistically significant (18.8% vs 6.3%, p=0.29).

In both techniques turbidity didn’t change from day 6 to day 33, with distinction in frequency of turbidity which was higher at step-back technique compared to crown-down technique (25% vs 6.3%).

In both obturation techniques using EndoRez, turbidity appeared already in first day, at step-back technique in three cases (18.8%) and one case (6.3%) in crown-down obturation technique. At step-back technique highest value of turbidity was achieved in day six, in six cases (25%), whereas at crown-down technique the turbidity highest value didn’t change since the first day, because turbidity remained at only one case (56.3%). The level of coronal microleakage from root canal system obturated with EndoRez statistically didn’t show any significant importance between step-back and crown down obturation techniques (t-proportional test=1.46, df=30,p=0.15) (tab 2 and graph 2).
Coronal Leakage of two Different Root Canal Sealers
Tringa Kelmendi, and et al

Table 2. Presentation of coronal microleakage from samples treated with step-back and crown-down techniques and sealed with EndoRez.

Table 3. Minimum, maximum, mean and standard deviation microleakage values of the subgroups.

Table 4. Mean leakage between subgroups instrumented with both techniques after obturation.

Discussion

The treatment of apical periodontitis consists of eliminating of a microbial infection in the root canal system\(^1\). In teeth with apical periodontitis, bacteria invade and colonize the entire root canal system and treatment is shifted toward the removal of microorganisms from the root canal system and prevention of reinfection\(^2\).

Cleaning and shaping of the root canal system is recognized as being an indispensable part of endodontic treatment\(^3\,4\).

In the present study, we evaluated the effect of instrumentation technique and two obturation sealers on the amount of microleakage using a bacterial method. The results of our study demonstrated that the two instrumentation technique (step-back vs crown-down) of preparation showed no significantly different levels of leakage.

Many of researchers studied only one instrumentation technique or the impact of canal size preparation on microleakage\(^5,6\).

For in vitro microleakage evaluation research extracted human teeth are used. The presence of bacteria in the canal or recontamination of the specimens during the study may impact on bacteria method. Autoclaving teeth does effect dentinal structure enough to compromise research\(^7\). Storing teeth in formalin influences the microleakage\(^8\). In comparison to other sterilization methods, Gigasept PA is a potentially safe alternative for sterilizing extracted teeth\(^9\).
Coronal leakage of microorganisms through root filling may be an important cause of failure in root canal therapy. This study showed that leakage occurred after loss of the coronal seal, as had previously been shown by other studies. Bacteria were used to evaluate the coronal leakage because this method is considered to have more biological and clinical relevance than the dye leakage test. Torabinejad et al., showed that highly motile bacteria (P. vulgaris) penetrated completely in 66 days, whereas nonmotile bacteria (S. epidermidis) penetrated in 30 days. E. faecalis was the bacteria of choice for the present study because it is generally associated with endodontic treatment failure. Endodontic sealers have an important role in root canal obturation because they complete the spaces that are not filled by gutta-percha.

The results of this study showed that after 33 days, EndoRez and Apexit Plus show similar sealing ability. Apexit showed the worst sealing ability compared to other sealers, in Miletic et al. study. A similar effect as Miletic et al. was reported by Wu et al., who found the greatest leakage through Sealapex, a Ca(OH)2-containing sealer. In the study of Eldeniz and Ørstavik (2009) many specimens of Apexit group resisted bacterial penetration up to 40 days, and also concluded that Apexit sealer resisted bacterial penetration for a longer period of time than EndoREZ. Sabyasachi S et al. (2010), showed no antimicrobial activity of resin based sealer and calcium hydroxide based sealer against E. faecalis.

Since EndoRez is a resinous sealer, better sealing qualities were expected. Our results agree with Ormiga et al.(2016) and Bortlini et al. (2010) that EndoRez sealer showed a better sealing ability of the root canal system. Contrary to our study, Leal et al. (2014) reported that Apexit Plus prevented leakage better than EndoREZ. According to Meidyawati et al. (2017), the sealing ability of epoxy resin sealer was better than mineral trioxide aggregate sealer.

Materials tested in the present investigation showed similar leakage. All sealers leak, definitely leakage can’t be the only criterion that determines the reliability of an endodontic sealer, but also biocompatibility, fracture resistance, antimicrobial properties and good handling characteristics.

Conclusions
The following might be concluded root canal preparation with rigid stainless steel hand files can equally have leakage just like rotary files with no significant difference between the two, Apexit Plus and EndoRez sealers have similar coronal sealing ability. Under the conditions of this in vitro study one should take into consideration, that this experiment was conducted in vitro with its inherent limitations. Therefore, further clinical investigations should be performed for definitive conclusions.

Declaration of Interest
The authors confirm that this article content has no conflict of interest.

Data Availability
The data set to support the findings of this study were supplied by Tringa Kelmendi and Co-authors under license and so cannot be made freely available. Requests for access to these data should be made to Tringa Kelmendi contact email tringa.kelmendi@uni-pr.edu.

References


