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Original article

Microleakage Assessment of Single Cone Gutta Percha Obturation Technique Using Different Types of Sealers

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ABSTRACT

This study was carried out to compare the coronal and apical seal of canals obturated with single cone technique using different types of epoxy resin root canal sealers. A total of sixty extracted human upper molars with fully formed and sound palatal roots were used in this study. The crown of each tooth was decoronated at cement-enamel junction and the palatal root canal was prepared using ProTaper gold rotary instruments. Sixty specimens were used and divided into 3 equal groups(n=20) for single cone obturation using two different types of epoxy resin sealers AH26 and vioseal sealers respectively and one control group without sealer. All the specimens were exposed varnish application followed by dye penetration test using 2% methylene blue dye then root sectioning. Samples were evaluated under a stereomicroscope to detect the linear measurement of the dye penetration from the apical constriction. The image transferred to a computer equipped with the image analysis software program where the dye penetration along the filling dentine interface was evaluated. The collected data was statistically analysis by using Graph Pad Instat software for windows. One-way ANOVA was done for compared results followed by Tukey's pair-wise. The results showed that, the difference in dye penetration microleakage means between groups was statistically significant as revealed by one-way ANOVA test ($p < 0.05$). Pair-wise Tukey's post-hoc test showed non-significant ($p>0.05$) difference between (AH26 and Vioseal sealers) groups. None of the tested root canal sealers could eliminate the dye infiltration or microleakage formation. Single cone gutta-percha combined with AH26 sealer exhibited less microleakage than vioseal sealer although there were no statically significant differences between them.

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INTRODUCTION

The complete obturation of the thoroughly cleaned and shaped root canal system and the creation of a fluid-tight hermetic seal is the ultimate clinical objective of the endodontic therapy, since its quality can affect the outcome of the root canal treatment [1,2]. The three-dimensional obturation of the root canal system is essential to entomb residual bacteria and preventing any bacterial recontamination that would lead to relapse of the apical periodontitis [3].

The phenomena of microleakage which is identified as a clinical undetectable passage of bacteria, fluids, molecules or ions between tooth and filling material has been known for more than 100 years [4]. It was reported that presence of such leakage has a great influence on the long-term success of endodontic therapy as it may cause many severe biological effects leading to recurrence of the pathology and failure of the root canal treatment [5]. Various obturation techniques based on different strategies have been introduced for the filling of root canals such as the lateral condensation, vertical compression, and thermoplasticized gutta-percha techniques [6]. It was reported that lateral condensation and warm vertical compaction show some disadvantages, such as: lack of gutta-percha homogeneity, high percentage of endodontic cement at the apical portion of the root and poor adaptation to the root canal walls [7]. To overcome these disadvantages, the single cone technique has been recently developed with the advancement of the rotary instrumentation systems. This technique uses larger master cones that best match the geometry of the nickel-titanium rotary [8]. This technique has become popular among endodontists over the years due to its greater adaptability to the characteristics of nickel-titanium (NiTi) rotary systems [9]. Furthermore, it was reported that the use of these cones with endodontic cement when the root canal is enlarged with rotary instruments, may promote the sealing of the root canal without the need for accessory cones or lateral condensation which on turn reduces the working time, allows easier and faster filling, and hence, causing less fatigue for both the patient and the operator [10]. In addition, it was reported that the combination of single cone and endodontic cement results in minimum applied pressure to the root canal walls and with a uniform mass that prevents failures observed among multiple cones [11].
Endodontic sealers play a critical role in providing an impervious seal to reduce leakage as they fill the irregularities and minor discrepancies between the root canal walls and core filling material [12]. However, inappropriate sealer coating may result in voids and permit bacterial microleakage that can potentially lead to treatment failure [13]. A variety of sealers based in different compositions have been used for this purpose [14]. At present, sealers based on epoxy resins provide very good physical properties and ensure adequate biological performance [15]. Leakage along root fillings may increase or decrease during the course of a long period after filling. Dissolution of sealer and the smear-layer may result in a rise in leakage, whereas gutta-percha swelling may result in diminished leakage [16]. Most sealers shrink during setting, leaving unwanted voids, and gradually dissolve [17]. Their sealing ability is also influenced by such physical properties as viscosity, flow, setting time, and film thickness [18]. Therefore, Leakage studies on the sealing properties of endodontic materials constitute an important area of research. The aim of this in vitro study was to compare the coronal and apical seal of canals obturated with single cone technique using different types of epoxy resin root canal sealers.

METHODS

Specimens' selection

Sixty freshly extracted human upper first and second molar teeth with fully formed and sound palatal roots were selected for this study. Radiographs and visual inspection under a dental microscope (magnified lenses 20x) were used to exclude those with immature apex, cracks, previous endodontic treatment, root caries or root resorption from the study. The teeth immersed in 5.25% NaOCl solution for 5 min. for disinfection. Subsequently, the samples cleaned of tissue remnants and calculus by ultrasonic scaler followed by rinsing under tap water and then stored in normal saline solution until the time of use.

Specimens' preparation

The crown of each tooth was decoronated using a high-speed handpiece with TC-11C diamond bur under continuous water spray at the cemento- enamel junction in order to ensure that all the specimens were relatively at the same length 11± 4mm. The palatal canal was explored with K-file #10 then working length (WL) determined by inserting a K-file# 15 into the canal until it was just visible at the apical foramen, and then the actual length of the canal was determined by subtraction of 1 mm from the first measurement. The working length (W. L) of each root was registered in a table drew in a plain paper. The root canals prepared using ProTaper gold rotary instruments installed on an electrical endodontic handpiece (Eighteeth E-Connect S) at speed and torque as recommended by manufacturer, respectively. Preparation carried out according to the manufacturer's recommendations using the crown-down technique starting by Sx file then S1, S2, F1 and finally by F2 file. A five new set of instruments was used for each group of teeth (each set prepare four canals). No instrument separation occurred during preparation of the specimens.
The canal irrigation was performed between each step of instrumentation with 5 ml. of freshly prepared solution 5% NaOCl carried up to the apical 3 mm with 27-gauge disposable plastic syringes then intra canal ultrasonic activation for 30 second and k-File #10 used for canal patency after each irrigation. Needle tip of the plastic syringe was placed passively into the canal. Following instrumentation, root canals irrigated with 1 ml EDTA 17% for 1min. followed by

5 ml 2.5% NaOCl. Finally, the root canals flushed with 3 ml of saline solution and dried with F2 protaper gold paper points.

Root canal obturation

The teeth were randomly divided into 3 groups, consisting of two experimental groups (n=20) according to the types of epoxy resin sealers used in the study and one control group (n=20) without sealer. Both sealers were mixed according to manufacture recommendation and carried out into the prepared canals according to each group using #25 Lentulo spiral rotated 3 mm away from the apex [19]. Group I: Canals were filled with single cone ProTaper gutta-percha size F2 which sealed with AH26 sealer (sealer A). Group II: Canals were filled with single cone ProTaper gutta-percha size F2which sealed with Vioseal sealer (sealer B). In both groups, the tip of each prefitted master cone was slightly coated with its respective sealer and inserted into the prepared canal using up-and-down pumping motion until reaching the full working length [20]. The excess gutta-percha removed with a gutta-percha cutter device and the remaining gutta-percha vertically compacted at the canal orifice. For group III control (n=20), all the samples were obturated with single gutta-percha cone size F2 without sealer to have maximum dye penetration. All specimens were lift four weeks in room temperature to allow complete setting of the sealers [21].

Microleakage testing procedure

Dye penetration test

The roots were then placed in 2% methylene blue dye (Supreme organization for drugs, Germany) for 24 hr. in incubator (37^{o}C). The teeth were then removed from the dye and excess of dye was washed off under running tap water to remove dye on external root surface. The teeth were dried, and the apical surface gently ground on a fine (400 grit) sand paper to remove superficially adhering dye.

Root preparation and sectioning

The roots were then sectioned vertically along the long axis to ensure that the sectioning process did not damage the inside of the canal. The sectioning was initially done with diamond bur (KG Sorensen, SP industrial, Brazil) operated on low-speed drill (ARATHON, SAE YANG CO., Korea) (Figure.1-I) to make a guide for the chisel and mallet to complete the sectioning till separation (Figure.1- II & III).

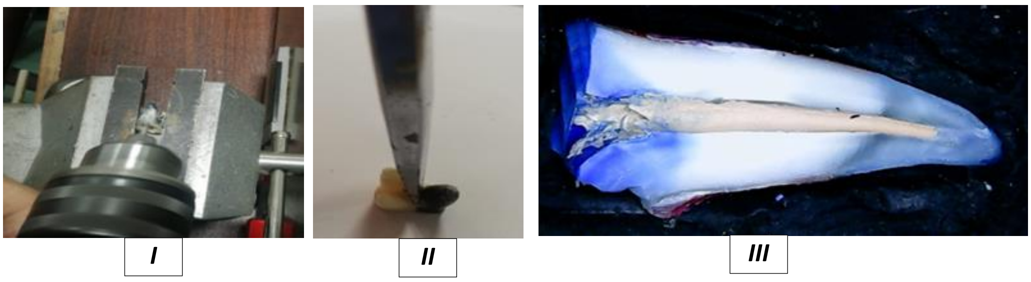

Figure 1. I). Root canal sectioning using kg Sorensen diamond bur. II). Using of a chisel and mallet to complete root sectioning till separation. III). Showing complete root section ready for microleakage evaluation

Microscopic evaluation

Samples were then evaluated under a stereomicroscope (Nikon Eclips E600, Tokyo, Japan) at ×25 magnification to detect the extent of dye penetration from the apical constriction, and the higher value among them was taken. With the help of the microscopic images obtained, the linear measurement of the dye penetration was noted from apical to coronal direction. The image transferred to a computer equipped with the image analysis software program (Image J 1.43U, National Institute of Health, USA), where the dye penetration along the filling dentine interface was evaluated (Figure.2).

Software calibration

Within the software, all limits, sizes, frames, and measured parameters are expressed in pixels. Therefore, system calibration was done to convert the pixels into absolute real-world units. Calibration was made by comparing an object of known size (a ruler in this study) with a scale generated by the software. Image J software was used to measure dye penetration expressed in mm.

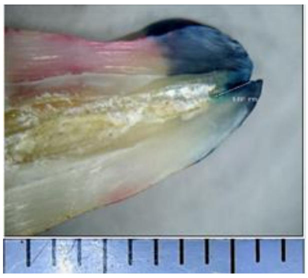

Figure 2. Image analysis software for evaluation of dye penetration leakage

Statistical analysis

The collected data was statistically analyzed by using Graph Pad Instat (Graph Pad, Inc., USA) software for windows. One-way analysis of variance (ANOVA) was done for compared results followed by Pair-wise Tukey's post-hoc test if showed significant results. The level of significance was set at of $P < 0.05$.

RESULTS

Microleakage results were measured by apico-coronal extension of dye in millimeter were summarized in (Table 1) and graphically drawn in (Figure.2, 3, 4, 5, 6).

According to (table1) which represented descriptive statistics of dye penetration leakage results for all the groups, the overall data showed that the experimental groups exhibited different degrees of dye leakage.

Our finding showed that the highest dye penetration leakage mean value was recorded for Control group (1.3±0.71 mm) followed by Sealer B group (0.54±0.23 mm) while the lowest dye penetration leakage mean value recorded for Sealer A group (0.50±0.29). The difference in dye penetration microleakage means between groups was statistically significant as revealed by one-way ANOVA test ($p < 0.05$). Pair-wise Tukey's post-hoc test showed non-significant ($p>0.05$) difference between (Sealer A and Sealer B) groups.

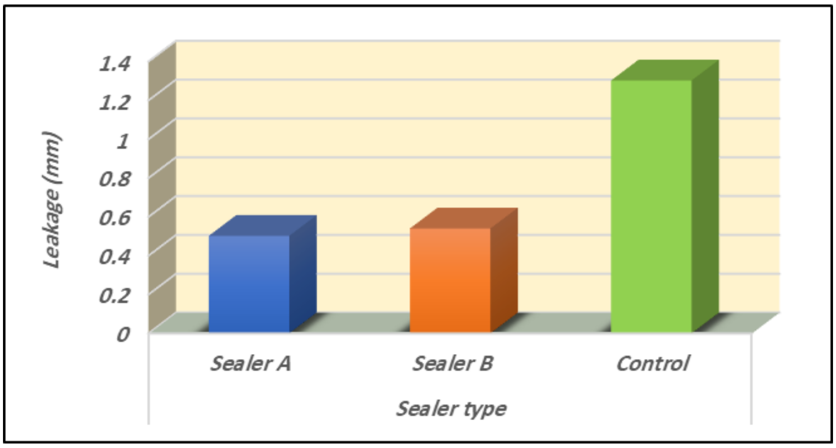

Figure 2. The means of dye penetration leakage for all groups.

Table 1. Descriptive statistics and Comparison of dye penetration leakage results (Mean ± SD) for all sealer groups (mm) using ANOVA and Tukey's Post hoc tests.

Variable		Descriptive statistics		Range		95% confidence intervals	
		Mean	SD	Minimum	Maximum	Low	High
Sealer type	Sealer A	0.50^{B}	0.29	0.04	1.64	0.37	0.62
	Sealer B	0.54^{B}	0.23	0.18	1.33	0.43	0.64
	Control	1.30^{A}	0.71	0.25	2.97	0.98	1.61
ANOVA	F	18.9					
	P value	<0.0001*					

*Different superscript capital letters indicating significant (p<0.05). ns; non-significant (p>0.05). *; significant (p<0.05)*

Figure 3. Stereomicroscopic image showing dye penetration leakage evaluation for sealer A group

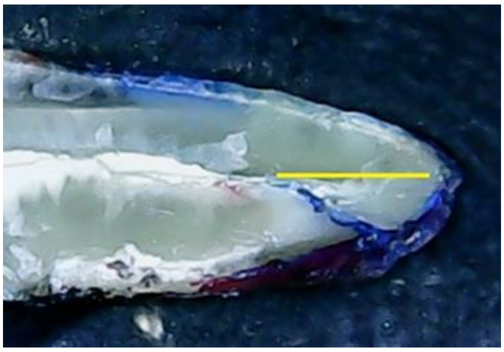

Figure 4. Stereomicroscopic image showing dye penetration leakage evaluation for sealer B group.

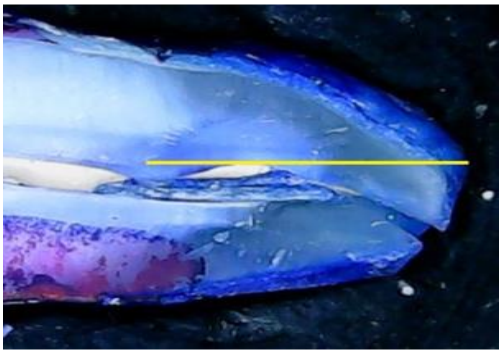

Figure 5. Stereomicroscopic image showing dye penetration leakage evaluation for Control group.

DISCUSSION

The success of a root canal therapy strongly depends on the thorough debridement of the root canal system, the elimination of pathogenic organisms and finally the complete sealing of the canal space to prevent ingress of bacteria from the oral environment and spread to the periapical tissue [20]. A three-dimensional obturation and creating a fluid-tight seal along all the communication pathways between the coronal and apical portions of a root canal system play a role in preventing percolation and microleakage of periapical exudate into the root canal space and create a favorable environment for healing to take place [22].

Root canal sealers based on epoxy resins were introduced in endodontic by Schroeder [4], and current modifications of the original formula are widely used for root canal filling procedures [23]. One of these sealers is (AH26) which has been extensively evaluated for its sealing ability and gap formation is characterized by a reactive epoxide ring and is polymerized by the breaking of this ring [24]. It was reported that AH26 showed good sealing ability, high tissue compatibility, it can harden in the presence of moisture, and it showed less than 0.5% restriction when entering the accessory canals [25]. AH26 has also been shown to have larger initial expansion compared to other types of epoxy – resin sealer [26]. However, the release of formaldehyde and its long setting time (4 weeks) are unfortunate the main disadvantages [22]. Another epoxy resin–based sealer is Vioseal which is a paste / paste type of dual syringe with very few reports in literature about its interfacial adaptation, leakage and void formations although the manufacturer claims that Vioseal has good sealing ability, superior biocompatibility, radiopacity, tight sealing, and insoluble in tissue fluids, excellent antibacterial effect and acts as a lubricant during inserting of GP point components [27].

The quality of apical seal obtained by root canal obturation material has been assessed by various methods like dye penetration, radioisotope penetration, bacterial leakage, fluorometric and electrochemical means, fluid filtration, scanning electron microscope and gas chromatography [28]. 2% methylene blue dye penetration method was selected as a leakage marker for the current study because; it is having a high degree of staining capability, a lower molecular weight and penetrates more deeply along the root canal filling [29-31]. Also, it easily allows quantitative measurement of the extent of the dye penetration by linear measurement techniques. Its molecular size is similar to bacterial by product such as butyric acid, which can leak out of infected root canals to irritate periapical tissues [32].

By going through the results of this study, it was observed that all the samples in the study group experienced apical microleakage. Apical microleakage is one of the initial causes of a root canal treatment failure, occurred due to inadequate obturation of root canal. Apical- coronal microleakage occurs on the surface of the sealer with dentin or with gutta-percha [33].

In the present study, the apical- coronal microleakage is characterized by the penetration of methylene blue as shown in table 1 and figure 2. The dye penetration was linearly measured from the apical end of root canal filling material toward its maximum coronal extent and not from the anatomical root end to assess the actual dye leakage alongside the obturation materials. The highest apical- coronal microleakage linear measurement was observed in the group 3 (control group) which had average value of 1.3 mm. followed by group II (sealer B) which had average value of 0.58 mm. and finally group I (sealer A) which had average value of 0.5 mm. The explanation of this finding could be attributed to several reasons.

For the control group which showed the highest level of dye penetration, indicated that sealing ability of single-cone gutta-percha is deteriorated when used without a root canal sealer since the beta type gutta-percha lacks adhesive quality as it was reported by Samiei et al., 2014 in their study [34]. Other explanation postulated that the gutta-percha serves as the core-filling material, whereas the root canal sealer forms a fluid tightly seal and barrier apically, laterally and coronally between the dentin and gutta-percha and that the sealer fills the space between gutta-percha cones depending on its adhesive properties [35].

For the epoxy resin-based sealer of the two study groups the apical microleakage occurs due to the natural properties of the sealer made from epoxy- resin, which is mainly due to the occurrence of shrinkage during polymerization which may cause bonding failure between the sealer and the root canal wall at first meeting as it was postulated by Wong and Cputo 2013 [36].

The result showed that although there were not statistically significance differences between the apical microleakage of the two sealer groups, AH-26 sealer (group A) demonstrated the least amount of microleakage. The results could be related to the considerable bond strength of AH-26 to dentin as well as gutta-percha because AH-26 mainly contains Bismuth oxide and hexamethylenetetramine as powder and Bis Phenol di-glycidyl ether as resin [24]. Also, the result could be attributed to the positive effect of EDTA on bonding of AH-26 sealer as it was postulated by Sheena et al., 2017 where smear layer removal by EDTA leads to expose dentinal tubules, creating rougher surface and producing greater adherence of AH-26 to the dentin through a micromechanical interlocking. Furthermore, the authors reported that the epoxy resin-based AH-26 sealer is thought to be able to react with any exposed amino groups in collagen to form chemical bonds between the resin and collagen when the epoxide ring opens. The potential of this chemical bonding due to ring opening explains the higher adaptation of AH-26 which is consistent in many studies [37]. Kokkas 2004 assessed the penetration depth of different sealers into the dentinal tubules and he found the penetration to be 10 to 80 μm after removal of the smear layer, whereas no penetration was observed with the smear layer intact [38]. Also, our results agreed with De Moore and De Bruyne 2004 study who assessed coronal and apical leakage via dye penetration of AH26 to other types of epoxy - resin sealers in 940 teeth obturated with different techniques. The authors reported that AH26 sealer produced the least microleakage among the sealer groups. Also, they reported that AH26 and other sealers resulted in comparable sealing ability at all evaluation times when used with identical obturation techniques [39].

Also, our results coincided with the results of Oliver and Abbott 1998 who found that AH-26 produced better the apical seal than the other tested groups of sealers which appeared to be due to bonding efficiently to gutta-percha than to dentine [40]. Our finding was coincided with the result of Sadr et al., 2015 in which, the positive control teeth showed maximum dye penetration and leaked at least 5 mm into the canals and AH-26 group had the least amount of micro-leakage compared to the other groups [41]. On the contrary of our result, Al-Anazi et al., 2020 reported in their study that vioseal sealer had a significant interfacial adaptation and least gap than other sealer groups due to its superior mechanical retention properties [42].

The discrepancy between the apical microleakage between the two sealer groups in our study could be also credited to the difference in density and diameter of dentinal tubules found at the apical level, resulting in lower sealer penetration [43]. Moreover, the smear layer removal is difficult at the apical third that might act as a physical barrier, which interfered with sealer adaptation to root canal dentin [44].

CONCLUSION

Within the limitations of this study and based on the results, it can be concluded that: none of the tested root canal sealer materials could eliminate the dye infiltration or microleakage formation. Single cone gutta-percha combined with AH26 sealer exhibited less microleakage than vioseal sealer although there were no statically significant differences between them.

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تقييم معدل الارتشاح لمادة حشوة قناة الجذر الفردية باستخدام عدة أنواع من مواد مانعات التسرب لحشو العصب

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المستخلص
تهدف هذه الدراسة الي مقارنة الختم الإكليلي والقمي للقنوات المسدودة بمواد سد مختلفة لقناة الجذر. تم استخدام ستين من الأضراس العلوية البشرية مع جذور حنكية مكتملة التكوين وسليمة في الدراسة. تمت إزالة تاج كل سن عند تقاطع المينا الأسمنتي وتم تحضير قناة الجذر الحنكي باستخدام أدوات بروتيبر جولد الدوارة. تم استخدام ستين عينة وقسمت إلى 3 مجموعات متساوية (ن = 20) لسد المخروط الفردي باستخدام نوعين مختلفين من السدادات راتنجات الايبوكسي أي إتش 26 والسدادات فايوسيل على التوالي ومجموعة تحكم واحدة بدون مادة مانعة للتسرب. تم تعريض جميع العينات بالورنيش متبوعًا باختبار الاختراق باستخدام صبغة زرقاء ميثيلين بنسبة 2 ٪ ثم تقطيع الجذر. ثم تم تقييم العينات تحت المجهر الضوئي لاكتشاف القياس الخطي لاختراق الصبغة من انقباض قمي. تم نقل الصورة إلى جهاز كمبيوتر مزود ببرنامج تحليل الصور حيث تم تقييم تغلغل الصبغة على طول واجهة عاج الأسنان. تم تحليل البيانات التي تم جمعها إحصائيًا باستخدام برنامج جراف باد إنستانت لنظام التشغيل ويندوز. تم إجراء أنوفا أحادي الاتجاه لوقت المقارنة متبوعًا بزوج توكي إذا أظهر نتائج تسريب كبيرة. أظهرت النتائج أنه بالنسبة لتقييم التسرب المجهري، كان الاختلاف في اختراق الصبغة يعني التسرب الدقيق بين المجموعات ذات دلالة إحصائية كما يتضح من اختبار أنوفا أحادي الاتجاه .($p <0.05$) أظهر اختبار زوج توكي فرقا غير مهم ($p> 0.05$) بين مجموعات (أي إتش 26 و فايوسيل). لا يمكن لأي من سدادات قناة الجذر المختبرة القضاء على تسلل الصبغة أو تكوين التسرب المجهري. أظهر جوتا بيركا المخروطي المفرد جنبًا إلى جنب مع مانع التسرب أي إتش 26 تسريبًا أقل من السداد الفيوسيل على الرغم من عدم وجود فروق ذات دلالة إحصائية بينهما.

الكلمات الدالة: أي إتش 26, فايوسيل، سدادات قناة الجذر، سد المخروط الفردي، معدل الارتشاح.