The effect of various endodontic irrigants on the sealing ability of Biodentine and other root perforation repair materials (In vitro study)

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ABSTRACT

Background: This in vitro study was carried out to evaluate the effect of various endodontic irrigants (sodium hypochlorite, ethylene diaminetetracetic acid and normal saline) on sealing ability of (Biodentine, mineral trioxide aggregate, and amalgam) used to repair furcal perforations.
Material and methods: One hundred and twenty extracted human molars with divergent roots were used in this study. A standard root canal access cavity was prepared in each tooth and furcal perforation was made and was standardized by using k file size 100 instrument to get a perforation of (1.32mm) in diameter .The teeth were randomly divided in to three groups of 40 teeth according to the type of material used to repair the perforations (Group A: The furcal perforations were repaired with Biodentine, Group B: The furcal perforations were repaired with MTA ,Group C: The furcal perforations were repaired with Amalgam). Each group was then subdivided into 4 subgroups according to irrigation regimens applied over the repair site (Subgroup 1: without irrigation, Subgroup 2: the pulp chamber was gently irrigated with 10 mL 5.25% Sodium hypochlorite for 10 minutes, Subgroup 3: Pulp chamber was gently irrigated with 10 mL 17% Ethylene Diaminetetracetic acid for 10 minutes, Subgroup 4: pulp chamber was gently irrigated with 10 mL normal saline for 10 minutes. Each tooth was coated with two layers of nail varnish and then sticky wax except 1 to 2 mm around the perforation site. Each tooth was placed in glass vial containing 3 ml of buffered Methylene blue dye at (37°C, pH 7) and kept in an incubator for 72 hour at 100% humidity. After dye application, the teeth were washed in running water for 5 min. Each tooth was sectioned longitudinally in a buccolingual direction.
Results: The results showed that group A has least mean of dye penetration and the difference was highly significant with group C and non-significant with group B.Saline and NaOCl increase the sealing of all groups while EDTA significantly increased the dye penetration of Biodentine and MTA respectively.
Conclusions: Biodentine has the best sealing ability of the tested materials while amalgam showed the highest dye penetration of all tested materials. Saline and NaOCl increase the sealing ability of Biodentine and MTA where as EDTA decreased the sealing efficacy of MTA and Biodentine.
Keywords: Biodentine, MTA, EDTA, NaOCl, root perforations. (J Bagh Coll Dentistry 2014; 26(3):1-8).

الخلاصة

الهدف من هذه الدراسة كان لتقييم تأثير مواد مختلفة لغسل قنوات الجذور (محلول الاثيلين دايمين تيترا استك اسد , محلول الهايبوكلورايد و المحلول الملحي العادي) على قدرة الختم لمواد (البايودنتين , مادة تجمع المعدن ثلاثي الاوكسد و الاملكم) المستعملة في ترميم ثقب مفترق الجذور .
استخدمت في هذه الدراسة مئة و عشرون ضرس سفلية حديثة القلع , متباعدة الجذور .تم تحضير قناه الجذر القياسية لكل سن ثم عمل انثقاب لمفترق الجذر و تم معايرتة بحيث يصبح بقطر (1,32 ملم) بواسطة استخدام مبرد k مقياس 100 , قسمت الاضراس عشوائيا الى ثلاث مجموعات تحوي على اربعين رحى استنادا لنوع المادة المستعملة في ترميم انثقاب مفترق الجذر كالاتي :
مجموعة أ : رممت انثقابات الجذور بمادة البايودنتين .
مجموعة ب : رممت انثقابات الجذور بمادة تجمع المعدن ثلاثي الاوكسد .
مجموعة ج : رممت انثقابات الجذور بمادة الاملغم
ثم تم تقسيم كل مجموعة إلى 4 مجموعات فرعية وفقا لنظم الري التي تم تطبيقها على موقع إصلاح مفترق الجذور على النحو التالي:
المجموعة الفرعية الاولى : لم يجري أي ري .
المجموعة الفرعية الثانية : تم ري غرفة اللب بلطف مع 10 مل من محلول الهايبوكلورايد 5.25٪ لمدة 10 دقائق .
المجموعة الفرعية الثالثة :تم ري غرفة اللب ب 10 مل من محلول الاثيلين دايمين تيترا استك اسد لمدة 10 دقائق
المجموعة الفرعية الرابعة : تم ري غرفة اللب ب 10 مل من محلول الملحي المتعادل لمدة 10 دقائق
تركت جميع الأسنان لتجف لمدة 24 ساعة ثم ملئت الاسنان بالحشوة المؤقتة. كل سن تم تغطيته بواسطة طبقتين من مادة طلاء الاظافر تليها طبقة من الشمع اللاصق باستثناء 1-2 ملم حول موقع انثقاب مفترق الجذور .
وضع كل سن في قارورة زجاجية تحتوي على 3 مل من صبغة الميثيلين الزرقاء مخزنة في (37 درجة مئوية، ودرجة الحموضة 7) و حفظ في حاضنة لمدة 72 ساعة وبعد تطبيق الصبغة، تم غسل الأسنان في المياه الجارية لمدة 5 دقائق . تم قطع كل رحى طوليا على طول المحور الطولي في الاتجاه الدهليزي اللساني من خلال الانثقاب باستخدام القرص الماسي . اظهرت النتائج ان المجموعة الاولى المرممة بمادة البايودنتين كان لها القيمة الوسطية النسبية الاوطا لاختراق الصبغة و كان الاختلاف ذو فرق معنوي احصائي عال جدا مع المجموعة الثالثة المرممة بواسطة مادة الاملكم و كان الفرق بين المجموعة الاولى و الثانية المرممة بمادة تجمع المعدن ثلاثي الاوكسد غير معنوي .
كما اظهرت النتائج ان المحلول الملحي العادي و و محلول الهايبوكلورايت يزيد قابلية الختم جميع المجموعات في حين محلول محلول الاثيلين دايمين تيترا استل اسد سبب في زيادة كبيرة في اختراق الصبغة من البايودنتين و مادة و تجمع المعدن ثلاثي الاوكسد على التوالي.

INTRODUCTION

An unfortunate but common complication of access preparation for endodontic therapy is a perforation made through the root into the surrounding periodontal tissues. The perforation normally occurs in the cervical area of anterior teeth, or in the furcation area of posterior teeth, as a result of the length of the bur being used [(1)].

Delay of perforation repair can cause microbial contamination of the defect and breakdown of the periodontium, resulting in endoperiodontal lesions that are difficult to manage. As such, perforation defects should be

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repaired before proceeding with definitive endodontic treatment [2].

Despite the numerous favorable properties of MTA that support its clinical use when compared with the traditional materials, there are several critical drawbacks such as the prolonged setting time, difficult handling characteristics, high cost, and potential of discoloration [3].

A variety of new calcium silicate–based materials have been developed recently aiming to improve MTA shortcomings. Biodentine (Septodont, Saint Maur des Foss_es, France) is a high-purity calcium silicate– based dental material that is recommended for use as a dentin substitute under resin composite restorations and an endodontic repair material because of its good sealing ability, high compressive strengths, short setting time,biocompatibility, bioactivity, and biomineralization properties [3],following the repair of furcal perforations, endodontic treatment should be performed with various irrigation solutions such as EDTA and NaOCl to clean the root canal system. This procedure causes inevitable contact of endodontic irrigants with the site of furcal repair in contrast to the well-documented chemical and morphological effects of routinely used irrigants (e.g., sodium hypochlorite [NaOCl] and ethylene diamine tetra-acetic acid [EDTA]) on root dentin and smear layer [4].

A little information exists with regard to the influence of these solutions on the integrity and sealing properties of repaired furcal perforations.The purpose of this in vitro study was to determine and compare the effect of various root canal irrigants (Sodium hypochlorite, Ethylene Diamine Tetraacitic Acid, and normal saline) on sealing ability of three different furcal perforation repair materials (Biodentine, Mineral trioxide aggregate, and amalgam).

MATERIALS AND METHODS

Selection of the samples

One hundred and twenty freshly extracted mandibular molars were used in the present study. The cause of extraction, age and gender were not considered. The molars were collected according to the following criteria: complete root formation, minimal occlusal or no caries, with diverged roots and no cracks on examination with 10X magnifying eye lens and light cure device [5,32].

After extraction, the teeth were cleaned by immersion in a 5.25% solution of sodium hypochlorite for 30 min. Soft tissue tags and calculus were removed by cumine scaler. Teeth were then washed with tap water and stored in normal saline until use [6].

To facilitate manipulation, the teeth were decoronated 3 mm coronal the cemento-enamel junction and the roots were amputated 3 mm apical the furcation using a diamond disk [2].

Preparation of the samples

All the teeth were prepared as following: a standard root canal access cavity was prepared in each tooth with a number 4 round bur in a high-speed handpiece with water spray for the initial entry followed by Endo-Z bur (long tapered configuration bur that allow easy access to the canal orifices and funnel shaping of the chamber walls) for lateral extension and finishing of cavity walls. The contents of the pulp chamber and root canals were removed with a spoon excavator and barbed broaches, respectively [2].

Creation of the Artificial Perforations

A heavy addition silicone impression material (Express STD 3M ESPE, Dental Products) was mixed according to the manufacturer instructions and placed in the area of artificial teeth of the manikin to provide a matrix that simulated the bony socket as it serves as a jaw for the teeth and was similar to barrier when condensing repair materials into perforation area.The molars were placed into the unset silicone impression material and then removed after complete polymerization of the material [7].

Artificial perforations were created in the center of the pulp chamber floor by using a size 2 round diamond bur (100 ISO size; Dentsply-Maillefer, Ballaigues, Switzerland) in a low-speed handpiece. The bur used for making a perforation was renewed every 5 samples. Then #100 K file was used to enlarge the perforation size so that the D16 facing the bottom of perforation to get standardized perforation diameter 1.32 mm at D16 of #100. The pulp chamber was filled with distal water for 3 minutes before being dried. Paper points were used to remove excess moisture within the perforation before repair [8].

For standardization, the heights of the walls of the perforated area in all the samples was 2mm which measured by using a periodontal probe. The samples in which the thickness of dentin at the perforated area was less than 2 mm were excluded [9].

Sample grouping

One hundred and twenty mandibular molars were randomly divided into three groups (n =40) according to the material used for repairing the perforation defects: (Group A:In which the perforations were repaired with Biodentine, Group B:In which the perforations were repaired with MTA, Group C: In which the perforations were repaired with amalgam). All the restorative materials used in this study were mixed according to the manufacturer's recommendations

For each restorative material, the specimens were further divided randomly into 4 groups (n=10 each) according to irrigation regimens applied over the repair site [10] (Subgroup 1: Without irrigation, Subgroup 2: the pulp chamber was gently irrigated with 10 mL 5.25% NaOCl and left for 10 minutes using disposable syringe, Subgroup 3: pulp chamber was gently irrigated with 10 mL 17% EDTA and left for 10 minutes using disposable syringe, Subgroup 4: pulp chamber was gently irrigated with 10 mL normal saline and left for 10 minutes using disposable syringe.

One investigator performed all procedures. In Subgroups 2, 3, and 4, the cavities were gently irrigated with 10 mL distilled water after irrigation of the tested solutions' to neutralize the prolonged effect of irrigants [2]. All the teeth were left to dry for 24 hour and all the access cavities were filled with temporary filling material.

Leakage study

Each tooth was coated with two layers of nail varnish except 1-2 mm around perforation and then sticky wax was melted in a cauldron and the roots were dipped in it by holding tooth with endodontic spreader to achieve a noticeable wax-covering of the apical and lateral portions of the roots, but avoiding wax coverage on the 1 to 2 mm around the perforation site and was left to dry.This will give complete sealing of the tooth [8].

After storage for 24 hours in incubator, each tooth was placed in glass vial containing 3 ml of Methylene blue dye at (37°C, & pH 7) and kept in incubator for 72 hour at 100% humidity.After dye application; the teeth were washed in running water for 5 min. The sticky wax and nail varnish were scrapped from the tooth with lacron carver and washed again under running water for 30 min. Then the teeth were left to dry at room temperature for 24 hours [8].

To evaluate the depth of dye penetration, each tooth was sectioned longitudinally in a in bucco-lingual direction, which was approximately parallel to the long axis of the tooth and through repaired perforation using diamond disk (0.3 thickness). Each half was fixed on a glass slide with sticky wax and each root given a special number so that data collection became easier [11].

Linear dye penetration was measured using a stereomicroscope with a 0.1-mm ocular grid at 20X magnification by using a millimeter grid digital vernier. The extension of dye penetration between the furcation filling material and tooth structure along bifurcation was assessed by two examiners calibrated for the technique and blinded to the groups. The measurement was made from bifurcation to the point where the dye no longer penetrated between the filling material and dentinal wall on both halves of each tooth. Accordingly, four leakage measurements were obtained for each specimen. The highest leakage values per specimen attributed by theexaminers were selected and micro-leakage means recorded for the experimental groups [11].

RESULTS

The results of the descriptive statistics which include the minimum, maximum, mean, and standard deviation values offurcal microleakage for three types of root perforation material using different types of root canal irrigants are shown in Table (1).

Table (1) and Figure (1) showed the following: the lowest mean value of furcal dye penetration in teeth repaired by Biodentine group (A) was found in subgroup A_4 while the highest mean value of furcal dye penetration was seen in subgroup A_3, the rest mean values for study subgroups were fluctuating between these values . The highest mean value of furcal dye penetration in MTA group (B) was seen in subgroup B_3 while the lowest mean value of furcal dye penetration was found in subgroup B_4.The lowest mean value of furcal dye penetration in teeth repaired with amalgam group (C) was found in subgroup C_4 while the highest mean value of furcal dye penetration was seen in subgroup C_3.

ANOVA test was performed to identify the presence of statistically significant differences for each type of material among different irrigant subgroup. The result showed highly significant differences in group A and group B regarding different irrigant solutions (NaOCl, EDTA and saline) while no significant differences were seen in group C as seen in Table (2)

The least significant difference test (LSD) was performed to evaluate the significant differences between different irrigant subgroup for each type of material and the result listed in Table (3). Table (3) showed that:

1. There was a significant difference between subgroup A1 and subgroup A2 while there was a highly significant difference between subgroup A1 and subgroup A3 and between subgroup A1 and subgroup A4.
2. There was highly significant difference between subgroup A2 and subgroup A3 and between subgroup A4 and subgroup A3 while no

significant difference between subgroup A2 and subgroup A4.

3. There was a highly significant difference between subgroup B1 in which teeth repaired with MTA without irrigation and subgroup B3 in which MTA irrigated with EDTA while there was significant difference between subgroup B1 and subgroup B2 when MTA irrigated with NaOCL and between subgroup B2 and subgroup B4 when MTA irrigated with normal saline.

4) There was a highly significant difference between subgroup B2 and subgroup B3 and between subgroup B4 and subgroup B3 while there were no significant difference between subgroupB2 and subgroup B4.

Table 1: Descriptive statistics

Groups	Subgroups	Mean	±S.D.
A (Biodentine)	**A1 (Without)**	0.37	0.08
	A2 (NaOCl)	0.28	0.08
	A3 (EDTA)	0.61	0.14
	A4 (Saline)	0.25	0.05
B (MTA)	**B1 (Without)**	0.42	0.09
	B2 (NaOCl)	0.37	0.08
	B3 (EDTA)	1.08	0.24
	B4 (Saline)	0.31	0.09
C (Amalgam)	**C1 (Without)**	1.71	0.26
	C2 (NaOCl)	1.65	0.23
	C3 (EDTA)	1.75	0.16
	C4 (Saline)	1.64	0.26

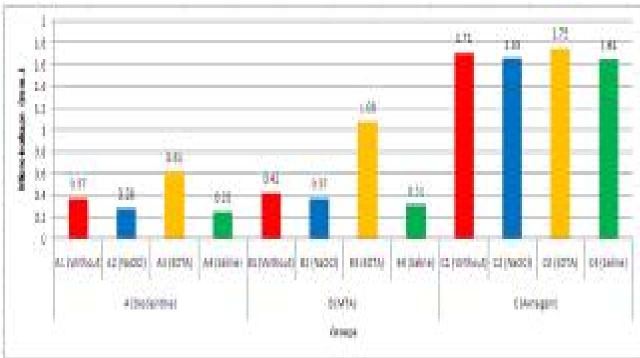

Figure 1: Mean of furcal dye penetration of different materials using different irrigants

Table 2: ANOVA test for furcal dye penetration among different irrigant within each type of material

Groups	Sum of Squares	Mean Square	F-test	p-value
A	0.8	0.27	30.8	0.000 (HS)
	0.31	0.01		
	1.11			
B	3.88	1.29	62.7	0.000 (HS)
	0.74	0.02		
	4.62			
C	0.08	0.03	0.5	0.682 (NS)
	1.92	0.05		
	2			

P≥ 0.05= N.S (Non-significant), P ≥ 0.05≥ 0.01 =S (significant), P≤ 0.01=Highly significant (HS)

Table 3: LSD test for furcal dye penetration among different irrigant for each type of material

Materials	Irrigants		Mean Difference	p-value
A (Biodentine)	**A1**	**A2**	0.09	0.037 (S)
		A3	-0.24	0.000 (HS)
		A4	0.12	0.007 (HS)
	A2	**A 3**	-0.33	0.000 (HS)
		A4	0.03	0.475 (NS)
	A3	**A4**	0.36	0.000 (HS)
B (MTA)	**B1**	**B2**	0.05	0.041 (S)
		B3	-0.66	0.000 (HS)
		B4	0.11	0.095 (S)
	B2	**B 3**	-0.71	0.000 (HS)
		B4	0.06	0.356 (NS)
	B3	**B4**	0.77	0.000 (HS)

Evaluation of furcal dye penetration among different group of materials for each type of irrigant was done using ANOVA test (Table 4) which revealed the presence of highly significant differences.

Least significant difference (LSD) test was performed to find source of significance Table (5). The result of LSD test showed: 1) There was a highly significant difference between group (A) in which perforations repaired by Biodentine and group (C) when amalgam used to seal the perforations and between group (B) in which perforation repaired with MTA and group (C) while that there were a no significant difference between group (A) and group (B) when no irrigation used. 2) There was a no significant difference between group (A) and group (B) while there were a highly significant difference between group (A) and group (C) and between group (B) and group (C) when NaOCl used. 3) There was a highly significant difference between all groups of material when EDTA used. 4) There was a highly significant difference between group (A) and group (C) and between group (B) and group (C) while that there were a no significant difference between group (A) and group (B) when normal saline used .

Table 4: ANOVA test for furcal dye penetration among different groups of materials within each type of irrigant

Irrigants	Sum of Squares	Mean Square	F-test	p-value
1 (Without irrigation)	11.54	5.77	208.85	0.000 (HS)
	0.75	0.03		
	12.29			
2 (NaOCl)	11.74	5.87	263.38	0.000 (HS)
	0.6	0.02		
	12.35			
3 (EDTA)	6.56	3.28	95.29	0.000 (HS)
	0.93	0.03		
	7.49			
4 (Normal saline)	12.35	6.17	238.84	0.000 (HS)
	0.7	0.03		
	13.05			

Table 5: LSD test for furcal dye penetration among different groups of material within each type of irrigant

Irrigants	Materials		Mean Difference	p-value
1 (Without irrigation)	A1	B1	-0.05	0.507 (NS)
		C1	-1.34	0.000 (HS)
	B1	C 1	-1.29	0.000 (HS)
2 (NaOCl)	A2	B2	-0.09	0.189 (NS)
		C2	-1.37	0.000 (HS)
	B2	C2	-1.28	0.000 (HS)
3 (EDTA)	A3	B3	-0.47	0.000 (HS)
		C3	-1.14	0.000 (HS)
	B3	C3	-0.67	0.000 (HS)
4 (Normal Saline)	A4	B4	-0.06	0.411 (NS)
		C4	-1.39	0.000 (HS)
	B4	C4	-1.33	0.000 (HS)

DISCUSSION

Perforations are procedural accidents that can have an adverse effect on the outcome of endodontic treatment. Identification of root perforations is possible by diagnostic aids that include direct observation of bleeding, indirect bleeding assessment using a paper point, radiography and an apex locator [(12)].

Dye penetration technique has been used in this study because of its ease of performance as compared to other available techniques. However, the dye penetration method is said to have certain drawbacks including the smaller molecular size of the dye molecules than bacteria, which do not measure the actual volume absorbed by the sample but merely measure the deepest point reached by the dye. It relies on randomly cutting the roots into two pieces, without any clue of the position of the deepest dye penetration [(13)].

Despite these drawbacks, a material that is able to prevent the penetration of small molecules (dye) should be able to prevent larger substances like bacteria and their byproducts [(12)]. Based on this, dye penetration seems to be a reliable technique and thus 2% Methylene blue dye was used in the present study.

Effects of different materials on sealing ability of furcal perforation:

The result of the present study showed the least mean value of the furcal dye penetration were found in Group (A) and when compared with other groups, there was highly significant difference between group (A) Biodentine and group (C) that used amalgam to repair perforation, while no significant difference between group (A) and group (B) that used MTA material .These results are in agreement with Koate and Pawar and Koubi et al. [(14,15)]. This could be attributed to: the superior ability of the calcium silicate materials to form hydroxyapatite crystals at the interface between the restorative material and the dentin walls, these crystals may contribute to the sealing efficiency of the material. Just after mixing, the calcium silicate particles of Biodentine, like all calcium silicate materials, react with water to form a high-pH solution containing Ca2+, OH−, and silicate ions. In the saturated layer, the calcium silicate hydrate gel precipitates on the cement particles, whereas calcium hydroxide nucleates [(16-18)].Also the small size of forming gels may contribute to better spreading of the material onto the surface and better fitting to dentin walls; a slight expansion of the calcium silicate-based materials may also explain the good sealing qualities of the calcium silicate cement [(19)].This finding disagree with the study done by Sanghavi et al.; this could be due to :1. Different method used by the researcher where a larger perforation (2mm) in width he made in his study while in this study the perforation width was (1.32 mm) in width which made it difficult to seal the defect effectively. 2. The researcher used ProRoot MTA while in this study, MTA angelus was used which has different composition. MTA-Angelus does not contain calcium sulfate and has lower percentage of bismuth oxide. This caused a reduction of the setting time from 2 hours for ProRoot MTA to 10 minutes for MTA-Angelus [(21)]

In this study the results also show that Group (B_1) in which perforations repaired with MTA have significantly higher difference than group (C_1) that used amalgam for repair this coincide with finding of [(14,22)]. This could be attributed to: Water based cements have been demonstrated good performance to seal the furcal perforation when compared to other materials. This moisture has favorable effect for MTA and Biodentine because of being water based materials has the ability to set in the presence of moisture while this moisture has a negative effect on amalgam which requires a dry field for good properties and that's difficult to obtain clinically [23].The main constituents of this material are calcium silicate, bismuth oxide, calcium carbonate, calcium sulfate, and calcium aluminate. Hydration of the powder produces a colloidal gel that solidifies into a hard structure consisting of discrete crystals in an amorphous matrix [24].

The result of the study also showed that amalgam has higher furcal dye penetration and there was a significant difference with group (A) and group (B) and this due to the defect were bottomless so the amalgam cant be well condensed to the perforation to provide the adequate seal and the amalgam is bonded mechanically and there is no chemical bonding between the amalgam and the opened dentinal tubules [25].

Effect of different endodontic irrigation solutions

1. Effects of EDTA

The results of this study indicated that there was a highly significant difference between subgroup A_3 and subgroup A_1 and subgroup B_3 and subgroup B_1. This finding is in accordance with other studies [2,26]. These results can be explained: 1) As pH decreases, the leakage of MTA increases owing to the acidic pH, the solubility of these repair materials may increase, which, in turn, might adversely affect their sealing ability. Moreover, calcium- depleting irrigants (EDTA) are capable of dissolving the smear layer rapidly (in this particular case, the smear layer on the inner surface of bur-cut perforation), and infiltrate into the interfacial layer; where they can interfere with the chemical adhesion between repair materials and dentin. Indeed, decomposition of particle-binding hydration phases by acid treatment raises potential concern on the strength and sealing properties of MTA-repaired perforations following irrigation by EDTA. As known, the hydration phases are responsible for the strength and barrier properties of MTA [27,28]. 2) Another explanation could be the demineralization effect of EDTA on Ca-containing materials. Because of the porous nature of MTA, it was conceivable that this precipitation proceeded internally within MTA to change the microstructure of MTA and thus led to a significant decrease in bond strengths of MTA-dentin which in turn reduce sealing ability [29,31].

The result also showed that there was a highly significant difference between subgroup A_3 in which perforation repaired with biodentine and subgroup B_3 in which MTA used to repair perforation. This finding agrees with Popali et al. This can be explained on the basis of calcium chloride present in the Biodentine liquid that's supplied by the manufacture .The addition of CaCL2 is intended to reduce the setting time and improve physiochemical properties by its ability to penetrate the pore of the cement, strongly accelerating the haydration of the silicate and leading to their faster crystallization [30].

Effects of NaOCl

The results of this study showed increase in the sealing of Biodentine when irrigated with NaOCL subgroup A_2 when compared with subgroup A_1 in which perforation repaired with Biodentine with no irrigation used and the difference were significant.This could be due to: 1)When Biodentine was exposed to NaOCl, it increased the size and amount of calcium hydroxide crystals compared with the control group, release of calcium, production of calcium hydroxide and increasing the PH all play apart in increase the sealing of the material [3]. 2) The biomineralization ability of Biodentine, most likely through the formation of tags, ion uptake into dentin leading to the formation of tag-like structures in Biodentine was higher than in MTA[3]. 3) NaOCl has an alkaline pH of 9.0 - 10.5, the literature indicated that high pH environments may enhance various physical and chemical properties of calcium silicate material [2].

The results also showed increase in the sealing of subgroupB_2 in which MTA where used for repair and irrigated with NaOCL when compared to subgroup B_1 in which no irrigation used but the difference were not significant. These results were in coincide with Uyanik et al. and this can be explained by: NaOCl is a halogenated compound that can cause mineral accumulation in human root dentin and expose inorganic material which may prevent dentin dissolution or may leave a smear layer of mineralized tissue that could increase the Calcium /Phosphate ratio of the dentin surface.

Effects of normal saline

The results of this study indicate that there were a highly significant difference between subgroup A_4 and subgroup A_1 in which no irrigation used; this could be attributed by: 1) When Biodentine exposed to saline one may speculate that additional unreacted mineral oxides may have remained that, once additional hydration was supplied, solidified and further increased the strength of the material. 2) Different chemical composition of Biodentine which contain calcium chloride in the liquid. The addition of $CaCL_2$ reduce the setting time and improve physiochemical properties by its ability to penetrate the pore of the cement, strongly accelerating the hydration of the silicate, reduce the incorporation of water, allow the cement to resist hydrostatic pressure, even at early stages, therefore avoiding the leakaging of cement and leading to their faster crystallization. When saline used to irrigate MTA repaired perforation, the sealing increased but the difference was significant when compared with control group in which no irrigation used.

This result agrees with other studies [1,3]; this may be due to of the remaining unreacted mineral oxides which may be solidified after additional supplied hydration and may result in the increased strength of the material.

REFERENCES

1. Loxley EC, Liewehr FR, Buxton TB, McPherson JC 3rd. The effect of various intracanal oxidizing agents on the push-out strength of various perforation repair materials. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2003; 95: 490–4.
2. Uyanik MO, Nagas E, Sahin C, Dagli F, Cehreli ZC. Effects of different irrigation regimens on the sealing properties of repaired furcal perforations. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2009; 107: e91–5.
3. Guneser MB, Akbulut MB, Eldeniz AU. Effect of various endodontic irrigants on the push-out bond strength of biodentine and conventional root perforation repair materials. J Endod 2013;39: 380-4
4. Torabinejad M, Cho Y, Khademi AA, Bakland LK, Shabahang S. The effect of various concentrations of sodium hypochlorite on the ability of MTAD to remove smear layer. J Endod 2003; 29: 233–9
5. Hashem A, Amin S. The effect of acidity on dislodgment resistance of mineral trioxide aggregate and bioaggregate in furcation perforations: an in vitro comparative study. J Endod 2012; 38(2): 245-9 .
6. Zou L, Liu J, Yin S, Li W, Xie J. In vitro evaluation of the sealing ability of MTA used for repair of furcation perforation with and without the use of internal matrix. Oral Surg Oral Med Oral Path Endod 2008;105(6): 61-5.
7. El-Tawil Sh, El-Dokky N, El-Hamid D. Sealing Ability of MTA versus Portland Cement in the Repair of Furcal Perforations of Primary Molars: A Dye Extraction Leakage Model. J Am Sci 2011; 7(12):1037-43
8. Alaajam W. Evaluation of the sealing abilities of different materials used to repair furcal perforations (in vitro study). A master thesis College of dentistry, university of Baghdad, 2002.
9. Rahimi S, Ghasemi N, Shahi S, Lotfi M, Froughreyhani M, Milani A, Bahari M. Effect of Blood Contamination on the Retention Characteristics of Two Endodontic Biomaterials in Simulated Furcation Perforations. J Endod 2013; 39(5): 697-700.
10. Zaparolli D, Saquy P, Cruz-Filho A. Effect of sodium hypochlorite and EDTA irrigation, individually and in alternation, on dentin microhardness at the furcation area of mandibular molars. Braz Dent J 2012; 23(6): 654-8.
11. Amin B, Dayem R, Salman R. The effect of two demineralizing agents on apical sealing of retrograde filling materials (In vitro study). Must Dent J 2009; 6(4): 299-306.
12. Reddy D, Kommineni N, Kumar, Hemadri M, Prasad S. Comparative evaluation of sealability of different root canal perforation repair materials by using a dye-extraction leakage method (An in virto study). Indian J Dental Sci 2013; 5(4): 44.
13. Camps J, Pashley DH. Reliability of the dye penetration studies. J Endod 2003; 29: 592– 4.
14. Kokate S, Pawar A. An in vitro comparative stereomicroscopic evaluation of marginal seal between MTA, glass inomer cement & biodentine as root end filling materials using 1% methylene blue as tracer. Endodontology 2012; 24(2): 36-42.
15. Koubi S, Elmerini H, Koubi G, Tassery H, Camps J. Quantitative evaluation by glucose diffusion of microleakage in aged calcium silicate-based open-sandwich restorations. Int J Dent 2012: 105863.
16. Pellenq RJM, Kushima A, Shahsavari R, et al. A realistic molecular model of cement hydrates. Proceedings of the National Academy of Sciences of the United States of America. 2009; 106(38): 16102-7.
17. Gandolfi MG, Van Landuyt K, Taddei P, Modena E, Van Meerbeek B, Prati C. Environmental scanning electron microscopy connected with energy dispersive X-ray analysis and Raman techniques to study Proroot mineral trioxide aggregate and calcium silicate cements in wet conditions and in real time. J Endod 2010; 36(5): 851–7.
18. kinner LB, Chae SR, Benmore CJ, Wenk HR, Monteiro PJM. Nanostructure of calcium silicate hydrates in cements. Physical Review Letters. 2010; 104(19): 195502.
19. Gandolfi MG, Ciapetti G, Perut F, Taddei P, Modena E, Rossi PL, Prati C. Biomimetic calcium-silicate cements aged in simulated body solutions. Osteoblast response and analyses of apatite coating. J Appl Biomater Biomech 2009; 7(3): 160–70.
20. Sanghavi T, Shah N, Shah R. Comparative analysis of sealing ability of Biodentin and calcium phosphate cement against mineral trioxide aggregate (MTA) as a furcal perforation repair material (An in vitro study). NJIRM 2013; 4(3): 56-60.
21. Sluyk SR, Moon PC, Hartwell GR. Evaluation of the setting properties and retention characteristics of mineral trioxide aggregate when used as a furcation perforation repair material. J Endod 1998; 24: 768-71.
22. Pereira CL, Cenci MS, Demarco FF. Sealing ability of MTA, Super EBA, Vitremer and amalgam as root-end filling materials. Braz Oral Res 2004; 18: 317–21.
23. Torabinejad M, Smith PW, Kettering JD, PittFord TR. Comparative investigation of marginal adaptation of MTA and other commonly used root end filling material. J Endod 1995; 21: 295-6.
24. Tsatsas DV, Meliou HA, Kerezoudis NP. Sealing effectiveness of materials used in furcation perforation in vitro. Int Dent J 2005; 55: 133-41.

25. Ahangari Z, Karami M. Evaluation of the sealing ability of amalgam, MTA, Portland cement and Coltozol in the repair of furcal perforations. Iran Endod J 2006; 1(2): 60–4.
26. Popali G, Jadhav S, Hedge V. Effects of Acid environment on surface microhardness of Biodentine. World J Dent 2013; 4(2): 100-2.
27. Roy CO, Jeansonne BG, Gerrets T. Effect of an acid environment on leakage of root-end filling materials. J Endod 2001; 27: 7-8.
28. Lee YL, Lin FH, Wang WH, Ritchie HH, Lan WH, Lin CP. Effects of EDTA on the hydration mechanism of mineral trioxide aggregate. J Dent Res 2007; 86: 534-8
29. Yan P, Peng B, Fan B, Fan M, Bian Z. The effects of sodium hypochlorite (5.25%), Chlorhexidine (2%), and Glyde File Prep on the bond strength of MTA-dentin. J Endod 2006; 32: 58-60.
30. Hong ST, Bae KS, Baek SH, Kum KY, Shon WJ, Lee W. Effects of root canal irrigants on the push-out strength and hydration behavior of accelerated mineral trioxide aggregate in its early setting phase. J Endod 2010; 36: 1995–9.
31. Al-Aubaydi A, Aziz A. Effect of EDTA on apical leakage of resin based root canal sealer. J Bagh College Dentistry 2010; 22 (4):10-4.
32. Al-Jaff A, Al-Azzawi H. Comparison of bond strength in different levels of post space of fiber-reinforced post luted with different resin cements. J Bagh Coll Dentistry 2011; 23(3): 1-5.