The effect of smear layer on push-out bond strength to dentin of Bioceramic sealer (In vitro study)

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ABSTRACT

Background: Sealers should demonstrate adhesive properties to dentin, decreasing the chance of endodontic treatment failure. Increased adhesive properties to dentin may lead to greater strength of the restored tooth, which may provide greater resistance to tooth fracture and clinical longevity of an endodontically treated tooth. The aim of this study was to evaluate the shear bond strength of Bioceramic iRoot SP sealer, AH plus sealer and Apexit plus sealer in absence or presence of smear layer using push out bond strength test.

Materials and Methods: Sixty straight single roots of the mandibular premolars were selected for this study. All canals were instrumented using ProTaper rotary instruments to achieve tapered canal walls, instrumentation was done with copious irrigation of 5.25% sodium hypochlorite. Roots were randomly divided into three groups according to the type of sealer used (twenty teeth for each group): Group A: Apexit plus + gutta percha, Group B: AH plus sealer + gutta percha, Group C: iRoot SP sealer + gutta percha .Then groups were subdivided according to types of final irrigation into two subgroups. Groups (A1, B1, and C1) were irrigated with 5 ml of 5.25% NaOCl for 1 minute while Groups (A2, B2, and C2), the smear layer was removed with 5 ml of 17% EDTA for 1 minute. All groups were rinsed with distilled water and then obturated with cold lateral condensation technique, the roots then stored in moist environment at 37°C for one week. The roots were embedded in clear acrylic resin and three horizontal sections were prepared at a thickness of 1 mm ±0.1 in the apical, middle and coronal parts of each root. The test specimens were subjected to the push-out test method using a Universal Test Machine that carried 1-mm, 0.5- mm and 0.3-mm plungers for coronal, middle and apical specimens, respectively. The loading speed was 0.5 mm/ min. The computer showed the higher bond force before dislodgment of the filling material. These forces were divided by the surface area to obtain the bond strength in MPa.

Results: The results showed that the bond strengths of iRoot SP and AH Plus were significantly higher than those of Apexit plus, but there was no significant difference between the bond strength of iRoot SP and AH Plus.In terms of root segments, the bond strengths in the middle specimens and the apical specimens were higher compared with the bond strengths in the coronal specimens.

Conclusion: The presence or absence of smear layer did not significantly affect the bond strength of Bioceramic filling materials.

Keywords: Bioceramic sealer, smear layer, push out test. (J Bagh Coll Dentistry 2013; 25(4):5-11).

INTRODUCTION

The introduction of bioceramic technology is considered a dramatic change in endodontic obturation. The introduced iRoot SP (Innovative Bioceramix, Vancouver, Canada) is a premixed, ready-to-use injectable and hydrophilic cement paste. It is composed of calcium phosphate, calcium silicate, calcium hydroxide, zirconium oxide, filler, and thickening agents. One of its advantages is its ability to form hydroxyapatite during the setting process and ultimately create a bond between dentinal wall and the sealer. It has been shown that iRoot SP is equivalent to AH Plus sealer in apical sealing ability, Furthermore, it was demonstrated that I Root SP was significantly less toxic than AH Plus [1,2]. Instrumentation of root canals produces a smear layer consisting of inorganic and organic components. The mechanical interlocking of the sealer plug inside the tubules following smear layer removal has been suggested to improve retention of the material, which might improve the sealing ability [3].

On the other hand, it has been shown that the bond strength of some sealer cements to dentin was better in the presence of smear layer [4]. Furthermore, because the smear layer contains moisture and might act as a coupling agent, thereby improving the adaptation of hydrophilic materials to the root canal wall. The removal of smear layer might have a negative effect on hydrophilic root canal sealers such as BC Sealer [5].

The push-out test provides a better evaluation of bonding strength than the conventional shear test; because when using the push-out test, fracture occurs parallel to the dentine–bonding interface, which makes it a true shear test for parallel-sided samples [6,7]. Interfacial strength and dislocation resistance between the root filling material and the intra-radicular dentine have been evaluated using thin-slice push-out tests [8-10].

MATERIALS AND METHODS

Sixty freshly extracted mandibular premolars with straight single roots and close apices were used in this study. The age of patients range between (18-48) years but the reason of extraction and gender was not considered. After extraction, all teeth were stored in 0.1% thymol solution at

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room temperature. Any periodontal remnants or soft tissues were removed by periodontal curette and root surfaces were verified with magnified eye lens (10X) and light cure device for any defects and cracks. After the length of root was determined by digital calliper and marker to 14 mm from apex to cemento-enamel junction, the root was sectioned perpendicular to its long axis by using diamond disc in a straight hand piece with water coolant to facilitate straight line access for canal preparation and filling procedure, also to eliminate the variables in access preparation and get flat reference point for measurement [11].

The pulpal tissue was removed by using barbed broach and copious amount irrigation of 5.25% NaOCl. The potency of canals was verified by insertion of No.15 K file into canal until it was visualised at apical foramen. The exact working length was established by subtracting 1mm from this measurement which is 13 mm. A Silicon rubber base (heavy-body) was mixed (base and catalyst) according to manufacturer instruction and inserted in plastic containers then the sectioned root was inserted inside the rubber base. Heavy body was left to set forming a small block to facilitate handling of the roots during instrumentation and obturation technique. The roots were instrumented by Rotary ProTaper (NiTi) system from SX-F3. All instrumentation was carried out according to manufacturer's instructions and completed in a crown-down manner using a gentle in-and-out motion. Instruments were withdrawn when resistance was felt and changed for the next instrument. The root was flooded with 5 ml of 5.25%NaOCl solution delivered with needle tip gauge 27 placed within apical third passively without bending and washed after each file**.**

Sample grouping

The roots were randomly divided into three groups (n=20) according to types of root canal sealer used**:**

Group A. Apexit plus root canal sealer obturation.

Group B. AH plus root canal sealer obturation.

Group C. I root sp root canal sealer obturation.

Then each group was subdivided into two subgroups (n=10) according to the method of final irrigation. Sub groups **A1, B1 and C1;** The roots canals were irrigated with 5 ml of 5.25% NaOCl for one minute and then irrigated with 5 ml of distilled water.Sub groups **A2, B2 and C2;** The root canals were irrigated with 5 ml of 17% of EDTA for one minute and 5 ml of 5.25%of NaOCl one minute and then irrigated with 5ml of distilled water. All groups were obturated by lateral condensation technique.

Group A: The samples were obturated with gutta percha and Apexit plus root canal sealer. The sealer was mixed according to the manufactures instructions by one press on handle of cartridge, equal amounts of base and activator were dispersed and mixed on clean and dry glass slab. The mixture had homogenous creamy consistency that stringed out when elevated with cement spatula over glass slab for one inch**.** Each canal was dried with paper point size F3. The K type master cone of gutta percha size 30 was adjusted to working length with tug back. Protaper absorbent paper point size F3 was dipped in sealer and coated the canal walls by counter clock wise rotation. The tip of master gutta percha cone was dipped into the sealer and inserted to correct full working length. The previously checked finger spreader size 20 was inserted between master cone and the canal wall using firm (apical only) pressure to within (1-2mm) from working length**.** The spreader was moved apically with a 180° clockwise-anticlockwise movement. The tapering of spreader was a mechanical force that laterally compresses and spread gutta percha creating space for additional accessory cones size 15 and 20. When the spreader did not inter more than 2mm ,excess gutta percha was removed with heated instrument to level 1mm higher than the coronal end of the root and vertically condensed with root canal plugger so gutta percha was obturated the entire canal up to canal terminus. The load applied during condensation ranged from 1.5-2 Kg determined by weight balance [12]**.**

Group B: The samples in this group were obturated with gutta percha and AH plus root canal sealer. AH plus sealer was mixed according to the manufactures instructions, by mixing equal amounts (1:1) of paste A and paste B on glass slab with spatula. The mixture had homogenous consistency that stringed out at least 1 inch when spatula was raised slowly from glass slab and then the canals were obturated with the same manner used for group A.

Group C: The samples in this group were obturated with gutta percha and iRoot sp sealer (Bioceramic sealer). After removing the syringe cap, attached an intra canal tip securely. Insert the tip of the syringe into apical third of root canal, filling the root canal while withdrawing the intra canal tip and then place gutta percha points inside the root canal then complete obturation in same manner in previous groups.

After obturating the teeth samples of all groups, the gutta-percha was removed at 1 mm below the orifice. Then the canal orifice was sealed with glass inomer cement as temporary filling to serve as a barrier to the ingress of fluids. All obturated roots of all groups were removed from impression material and wrapped in saline moistened gauze in closed plastic vial allowing the sealer to set for 7 days at 37°C in an incubator [13,14].

Then the roots were embedded in clear acrylic resin [15]. Metal frame of (length 70mm, width 60mm and height 30mm) containing three cylindrical holes of (diameter 12 mm * height 25mm) was used into which the prepared acrylic was loaded. Before loading the mold with acrylic, the coronal end of the roots was fixed on the face of the end rod of the dental surveyor with a sticky wax. With the aid of dental surveyor the roots were centrally located within the acrylic blocks to ensure that the sectioning would be perpendicular to the long axis of the roots.The acrylic was prepared by mixing powder and liquid in a porcelain jar. The material was left undisturbed for few minutes until it reached the workable stage and loaded into the metal mold, the rod of the surveyor with the root fixed on its face was pushed into the acrylic with gentle pressure to allow the complete embedding of the root into the acrylic and to allow the escape of the excess material. The metal frame was taken from surveyor and the material was allowed to cure under cool water at 20°C, which was necessary to compensate for the anticipated rise in the temperature of the samples subsequent to the exothermic curing reaction of the cold cure resin. The acrylic blocks were allowed to cure completely for at least 30min as recommended by the manufacturers [16]. Root sectioning was done After complete curing of the acrylic mold, the metal mold was open. The excess acrylic was cut off using diamond disk mounted on straight hand piece and engine with a rotation speed regulator, the hand piece was fixed in a cutting device. The root was cut horizontally with flow cold water (19-25°C) to minimize smearing [16]. To get three sections of 1mm in thickness coronal, middle and apical, the cuts were made at 2,6,9 mm from coronal reference point respectively.

Push-out test was performed by applying a compressive load to the apical aspect of each slice via a cylindrical plunger mounted on Tinius-Olsen Universal Testing Machine managed by computer software. Samples were examined under the Nikon metallurgical microscope (magnification 50X) and pictures of both sides of each section are taken with digital camera which was connected with microscope, and measurements calculated using LUCIA G software analysis program . The obturated area of the section at each level was measured from the apical side to determine the size of punch pin [17]. Three different sizes of punch pins were used, 1 mm, 0.6mm, and 0.3mm diameter for the coronal, middle and apical slices respectively. The punch pins should provide almost complete coverage over the main cone without touching the canal walls and sealer [13,17]. The root filling in each section subjected to loading using a universal testing machine (WDW50) at a speed of 0.5 mm / min in an apical-coronal direction until the first dislodgment of obturating material and a sudden drop along the load deflection. The maximum failure load was recorded in Newton (N) and was used to calculate the push-out bond strength in mega-pascals (MPa) according to the following formula [18]**:**

$$(MPa) - \frac{\text{maximum load (N)}}{\text{adhesive area of root canal filling mm}^{\wedge}2}$$

The adhesion (bonding) surface area of each section was calculated as:

$(\pi r1 + \pi r2)$ *L. L was calculated as

$$L = \sqrt{(r1 - r2)2 + h^{\wedge}2}$$

π = 3.14; r1 = coronal radius, in mm; r2 = apical radius, in mm; h = thickness of section in mm, L = adhesion area.

ANOVA and Student t-test were performed as statistical analysis for push-out bond strength.

RESULTS

Mean values of push-out bond strength & standard deviations for all groups presented in (table 1). Both the highest and the lowest mean values for sealer push-out bond strength were seen at apical level of iRoot sp sealer without smear layer group C**2** (**4.889**) and middle level of Apexit plus sealer without smear layer group A**2** (**1.125**) respectively. The rest mean values for study groups were fluctuating between these values. To compare among the six groups systems at each level, ANOVA test was preformed to identify the presence of statistically significant differences for sealer push-out bond strength among different groups within each level. Highly significant differences were found in A**1**, B**1**, B**2**, and C**2** at all levels while significant difference was shown in A**2** while no significant difference was seen in C**1** Table (2).

The least significance difference test **(**LSD**)** was performed to evaluate the significant differences between six groups at each level and the results listed in Table (3) and showed the followings:

- Highly significant differences between three levels in Group A**1** (Apexit plus sealer with smear layer).

- Highly significant difference between coronal and middle level while no significant differences between coronal and apical, and between middle and apical levels in Group A**2** (Apexit plus sealer without smear layer).
- Significant differences between coronal and middle, and between middle and apical levels while highly significant differences between coronal and apical levels in Group B**1** (AH plus sealer with smear layer).
- No significant differences between coronal and middle, and between middle and apical levels while highly significant differences shown between coronal and apical levels in Group B**2** (AH plus sealer without smear layer).
- No significant differences between coronal and middle, and between middle and apical levels while significant differences appear between coronal and apical levels in Group C**1** (iRoot sp sealer with smear layer).
- Highly significant differences between coronal and middle and between coronal and apical levels while no significant differences between middle and apical levels in Group C**2** (iRoot sp sealer without smear layer).

Student t test showed:

- No significant differences in push out bond strength at middle and apical levels in presence or absence smear layer of apexit plus sealer except at coronal level highly significant difference.
- No significant differences in push out bond strength at all levels in presence or absence smear layer of AH plus sealer.
- No significant differences in push out bond strength at coronal and apical levels in presence or absence smear layer of iRoot sp sealer except at middle level significant difference.

DISCUSSION

The adhesive strength of root canal sealers has been examined by various methods that include shear bond strength, microtensile bond strength, and pushout bond strength testing. The push-out test is easy to reproduce and interpret and provides a realistic assessment of bond strength to dentin even at low levels [13].

Effect of Sealer Type on Bond Strength

The result of present study showed the highest mean value of push –out bond strength in Group **C2** that used iRoot sp sealer with removed smear layer (Table 1) and when compared with other sealers with same method of irrigation, there was very highly significant difference between group **C2** iRoot sp sealer and group **A2** Apexit plus sealer, significant difference between group **C2** and group **B2** AH plus sealer. These results are in agreement with the results of other studies [19,20], which conducted to evaluate and compare the fracture resistance of roots obturated with various contemporary canal-filling systems and it was concluded that the innovative bioceramic-based sealer (iRoot SP) may have the potentiality to strengthen endodontically treated teeth to a level comparable to that of intact teeth. This could be attributed to the nature of iRoot sp sealer being a true self adhesive material that would form a chemical and mechanical bond with dentin through the production of hydroxyapatite during setting when the material is exposed to a moist environment as that present within the dentinal tubules. In addition, the bioceramic sealer is hydrophilic, possessing a low-contact angle that would allow the sealer to spread easily over the canal wall providing adaptation and good hermetic seal through mechanical interlocking. In addition, the extremely fine particle size and the optimal premixed consistency introduced with a capillary tip introductory system might have enhanced its penetration to the full length of the canal. Furthermore, zirconium oxide, one of the constituents of the iRoot SP sealer, has been reported to possess high fracture toughness, tensile strength, and lower Young's modulus [1,19,20]**.** The result of this study disagreed with [14,22] who found that there was no significant difference between AH plus and Bioceramic sealer used with gutta percha, this may be related to difference in method of obturation, [22] in his study the canal was obturated by single cone technique in addition a slice thickness used in push out bond strength was 2mm while [14] measured only the push out bond strength of AH plus and Bioceramic sealer at middle third.

In this study when the smear layer left at the apical area, group **B1** AH Plus and group **C1** iRoot sp showed a significantly higher values than group **A1** Apexit plus, but there was no significant difference between **B1** AH Plus and **C1** iRoot sp groups, this coincide with the findings of [14,22-24] no significant difference between BC sealer and AH plus push out bond strength. The high bond strength of AH Plus may be explained by the formation of a covalent bond by an open epoxide ring to any exposed amino groups in collagen [25]**.** Other investigations have shown a high-quality properties with epoxy resin–based sealers, including very low shrinkage while setting, long-term dimensional stability, flow, and long setting time, AH Plus sealer penetrates deeper into the surface micro-irregularities [26]**.**

Effect of Root Section Level on Bond Strength

The push-out bond strengths in the middle and apical specimens were significantly higher than those of the coronal specimens in group **C1** iRoot sp sealer and highly significant in group **C2**. There were no significant differences between the push-out bond strengths in the middle and apical specimens in both groups **C1** and **C2**. The result of this study is aligned with [24] who assessed the push-out bond strength of two new calcium silicate-based endodontic sealers in the root canals of extracted teeth iRoot sp and MTA Fillapex sealers. Three horizontal sections were prepared coronal, middle and apical. Their result showed that there were no significant differences between the bond strengths in the middle and apical slices. Some authors have reported the bond strengths of different sealer to dentin were higher in the apical one-third than in the coronal third [27-29]**.** The higher bond strengths in the middle and apical specimens could be related to deeper sealer penetrations because of higher lateral condensation forces in apical third than coronal third and also could be a result of irregular dentine and devoid of tubules in apical part of roots which increase surface area of adhesion [24,30].

Effect of Smear Layer Removal on Bond Strength

This study showed that there were no significant differences in push out bond strength at coronal and apical levels in presence or absence of smear layer in iRoot sp sealer groups except at middle level a significant difference. This result agree with the result of other studies [14,31]**.**

The open tubules and the absence of smear layer do not improve adhesion of endodontic sealers. The authors suggest that perhaps the open tubules increase the stress at the sealer dentin interface and that the calcium and phosphate-rich smear layer and plugs are potential sites of sealer adhesion [31]. **Shokouhinejad et al.** [14] compared the bond strength of a new bioceramic sealer (EndoSequence BC Sealer) and AH Plus in the presence or absence of smear layer, and they concluded that the presence or absence of smear layer did not affect the bond strength of EndoSequence BC Sealer. They explained that it may be due to Bioceramic sealer includes a similar composition to white MTA [1], and some studies revealed that removal of the smear layer caused significantly more microleakage in the root canals and root end cavities filled with MTA [5,32].So within the limitation of this in vitro study, can be concluded the presence or absence of smear layer did not significantly affect the bond strength of filling materials, the bond strengths of IRoot SP and AH Plus were significantly higher than those of Apexit plus, no significant difference between AH Plus and I Root SP groups in the presence of smear layer at the apical specimens and in terms of root segments, the bond strengths in the middle specimens and the apical specimens were higher compared with the bond strengths in the coronal specimens.

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Table 1: Mean values of push-out bond strength and standard deviations at three levels in MPa.

Group	Level	Mean	±SD
A1	Coronal	2.8892	0.085815
	Middle	1.2567	0.31708
	Apical	1.734	0.406251
A2	Coronal	1.7935	0.49572
	Middle	1.1251	0.607396
	Apical	1.2975	0.548605
B1	Coronal	2.9464	0.521307
	Middle	3.5701	0.717203
	Apical	4.1697	0.629287
B2	Coronal	2.9971	0.836391
	Middle	3.4908	0.707482
	Apical	4.184	0.773615
C1	Coronal	3.4071	0.652522
	Middle	3.8086	0.75308
	Apical	4.2111	0.877046
C2	Coronal	3.5704	0.63928
	Middle	4.488	0.526285
	Apical	4.8891	0.598721

Table 2: ANOVA test for push-out bond strength among three levels for each type of sealer

Groups	Areas	Areas difference (d.f.=29)		
		F-test	P-value	Sig.
A1	Coronal Middle Apical	77.44	0.000	H.S.
A2	Coronal Middle Apical	3.95	0.03	S.
B1	Coronal Middle Apical	9.49	0.001	H.S.
B2	Coronal Middle Apical	5.91	0.007	H.S.
C1	Coronal Middle Apical	2.75	0.08	N.S.
C2	Coronal Middle Apical	13.71	0.000	H.S.

P ≥ 0.05: Non significant (NS) P < 0.05: Significant (S) P ≤ 0.01: Highly significant (HS)

Table 3: Student t test of push out bond strength between two levels of each group of sealer

Groups	Levels	t-test	P-value	Sig.
A	Coronal A1 Coronal A2	6.89	0.000	H.S
	Middle A1 Middle A2	0.61	0.55	N.S.
	Apical A1 Apical A2	2.02	0.06	N.S.
B	Coronal B1 Coronal B2	-0.16	0.87	N.S.
	Middle B1 Middle B2	0.29	0.78	N.S.
	Apical B1 Apical B2	-0.05	0.96	N.S.
C	Coronal C1 Coronal C2	-0.58	0.57	N.S.
	Middle C1 Middle C2	-2.34	0.03	S.
	Apical C1 Apical C2	-2.02	0.06	N.S.

P ≥ 0.05: Non significant (NS) P < 0.05: Significant (S) P ≤ 0.01: Highly significant (HS)