Original Article

Comparison of sealing ability of bioactive bone cement, mineral trioxide aggregate and Super EBA as furcation repair materials: A dye extraction study

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Abstract

Context: Sealing ability of furcation repair material.

Aims: To evaluate the sealing ability of bioactive bone cement, mineral trioxide aggregate (MTA) and Super Ethoxybenzoic Acid (EBA) as furcation repair materials in mandibular molars using a dye extraction leakage model.

Settings and Design: *In vitro*, dye extraction study.

Materials and Methods: Forty mandibular molars were randomly divided according to the material used to repair perforation: Group I-MTA, Group II-bioactive bone cement, Group III-Super EBA, Group IV-Control (furcation left unrepaired). All samples were subject to ortho grade and retrograde methylene blue dye challenge followed by dye extraction with 65% nitric acid. Samples were then analyzed using Ultra violet (UV) Visible Spectrophotometer.

Statistical Analysis Used: One way analysis of variance (ANOVA), Tukey-Kramer Multiple Comparisons Test.

Results: MTA and bioactive bone cement showed almost similar and lower absorbance values in comparison to Super EBA.

Conclusions: Bioactive bone cement provi ded an excellent seal for furcal perforation repair and at the same time it provided comfortable handling properties, which could overcome the potential disadvantages as faced with MTA.

Keywords: Bioactive bone cement; dye extraction study; furcation repair; mineral trioxide aggregate; spectrophotometer

INTRODUCTION

Today in the era of regenerative endodontics, continual research into the field of bio-materials, has made restoring back original form and function of even the most complicated cases a reality. A furcation perforation is one such complication that refers to mid-curvature opening in to the periodontal ligament space and leads to worst possible treatment outcome.[1]

Ideally, to prevent bacterial contamination, perforations should be repaired as quickly as possible with a biocompatible material. The most commonly used repair materials are amalgam, zinc oxide eugenol cement, calcium hydroxide, gutta-percha, glass ionomer cement, resin modified glass ionomer cements, intermediate restorative material (IRM), composite resin, Super EBA ethoxy benzoic acid and mineral trioxide aggregate (MTA).

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Bone cement is a potentially new repair material that has been investigated in dentistry recently as a root end filling material, but has been in use in oral and orthopedic surgery for past 40 years. Bone cements have many characteristics that make it well suited as a repair material for a variety of endodontic treatments. Good strength and load bearing capacity,[2,3] good handling and working properties, faster setting times of around 15 min,[4,5] tolerates a moist environment very well, good marginal adaptation as a root end filling material,[4] low cytotoxicity comparable to MTA.[5] However, bone cement does not show bioactivity, that is an essential property for repair materials. Various studies have shown that it is possible to induce bioactivity by modifying bone cement with a bioactive material like amorphous calcium phosphate, hydroxyapatite (HA), and tetracalcium phosphate, bioactive glass etc.[6]

In this *in vitro* study, preparation of a bioactive bone cement has been attempted by modifying the polymer/powder

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part of bone cement with MTA such that all the favorable properties of bone cement are retained and the potential disadvantages faced with MTA are overcome. The purpose of the study was to evaluate the sealing ability of MTA, Bioactive bone cement and Super EBA as furcation repair materials in mandibular molars using a dye extraction leakage model.

MATERIALS AND METHODS

Forty extracted intact, non-carious human mandibular molars with non-fused and well developed roots were used in this study. The teeth were stored in 0.2% sodium azide until further use. Molars were decoronated 3 mm above the cemento-enamel junction and roots were amputated 3 mm below the furcation. A standardized endodontic access opening was prepared in all 40 samples. Sticky wax was placed over the orifice of each canal, and the sectioned root surface including the pulpal floor. It was then coated with two layers of resin varnish (D-tech).

To ensure each perforation was centered between the roots, a black marker pen was used to mark the location of the defect. A defect 1 mm in diameter [Figure 1a and b] was made from the external surface of the tooth with a number 2 round carbide bur mounted on a high-speed hand-piece with air water coolant. The chamber and perforation were flushed with water and dried. The teeth were kept in an incubator at 37°C for 24 h simulating clinical conditions.

Perforation repair

They were then randomly divided into four groups with 10 samples each and perforations sealed. Group I and III were repaired with MTA and Super EBA (mixed according to manufacturer's instructions). Group IV, which was the positive control was left unrepaired. Group II was repaired with bioactive bone cement [Figure 1c].

Preparation of bioactive bone cement

Preliminary studies were undertaken to determine the optimum concentration of MTA and silane coupling agent (methacryloxypropyltrimethoxysilane (MPS) required for modifying bone cement without altering its handling properties.

Modification of powder

Mass ratio of 0.4 mg MTA was added to 0.6 mg bone cement (60:40) and was mixed together until all the MTA particles were miscible in to the polymer powder.

Modification of liquid

To 1 mL of monomer liquid, 1 drop of silane coupling agent (Monobond-S) was added and mixed together. The prepared powder and liquid of the modified bone cement was mixed in the ratio of 2:1 under ambient conditions at room temperature. Dough like consistency of cement, which was obtained after mixing was placed with a plastic instrument into the defect and compacted flush to the pulpal floor with condensers. Moist cotton pellets were placed passively between the roots in the furcation area and the teeth were kept in an incubator at 37°C for 24 h simulating clinical conditions

Dye extraction micro leakage evaluation

Each group was placed in separate petri dishes containing 2% methylene blue such that all the teeth were immersed in dye up to the cementoenamel junction for retrograde dye challenge [Figure 1d] and dye was added to access chamber of each teeth so that it was filled for orthograde dye challenge [Figure 1e]. All samples were stored similarly for 48 h.

After removal from the dye, teeth were rinsed under tap water for 30 min and varnish removed with a polishing disc. Each tooth was stored in a vial containing 1000 µl of concentrated (65 weight %) nitric acid for 3 days. The solutions thus obtained were centrifuged at 3500 rpm for 5 min.100 µl of the supernatant was then analyzed in a UV ultraviolet-visible spectrophotometer at 550 nm wavelength with concentrated nitric acid as the blank and readings were recorded as absorbance units. The obtained readings were statistically analyzed using one way ANOVA and Tukey multiple comparison tests.

RESULTS

The mean absorbance values [Figure 2] of experimental groups and controls in the current study [Tables 1 and 2] showed that the positive control samples (Group IV) in which perforations were left unrepaired had the highest dye absorbance (0.87 ± 0.09487) of all groups denoting the accuracy of the technique. This was followed by Super EBA (Group III) which had dye absorbance values (0.49 ± 0.07379) significantly higher than MTA (Group I) and

Table 1: Sphectrometric readings of individual groups

Groups	Mean sphectrometeic values (absorbauce units-AU)	Standard deviation	Standard error of mean	Median	95% confidence interval	
					From	To
MTA	0.1900	0.07379	0.02333	0.2000	0.1372	0.2428
BBC	0.2100	0.07379	0.02333	0.2000	0.1572	0.2628
Super EBA	0.4900	0.07379	0.02333	0.5000	0.4372	0.5428
Control	0.8700	0.09487	0.03000	0.9000	0.8021	0.9379

MTA: Mineral trioxide aggregate, BBC: Bioactive bone cement, EBA: Ethoxybenzoic acid

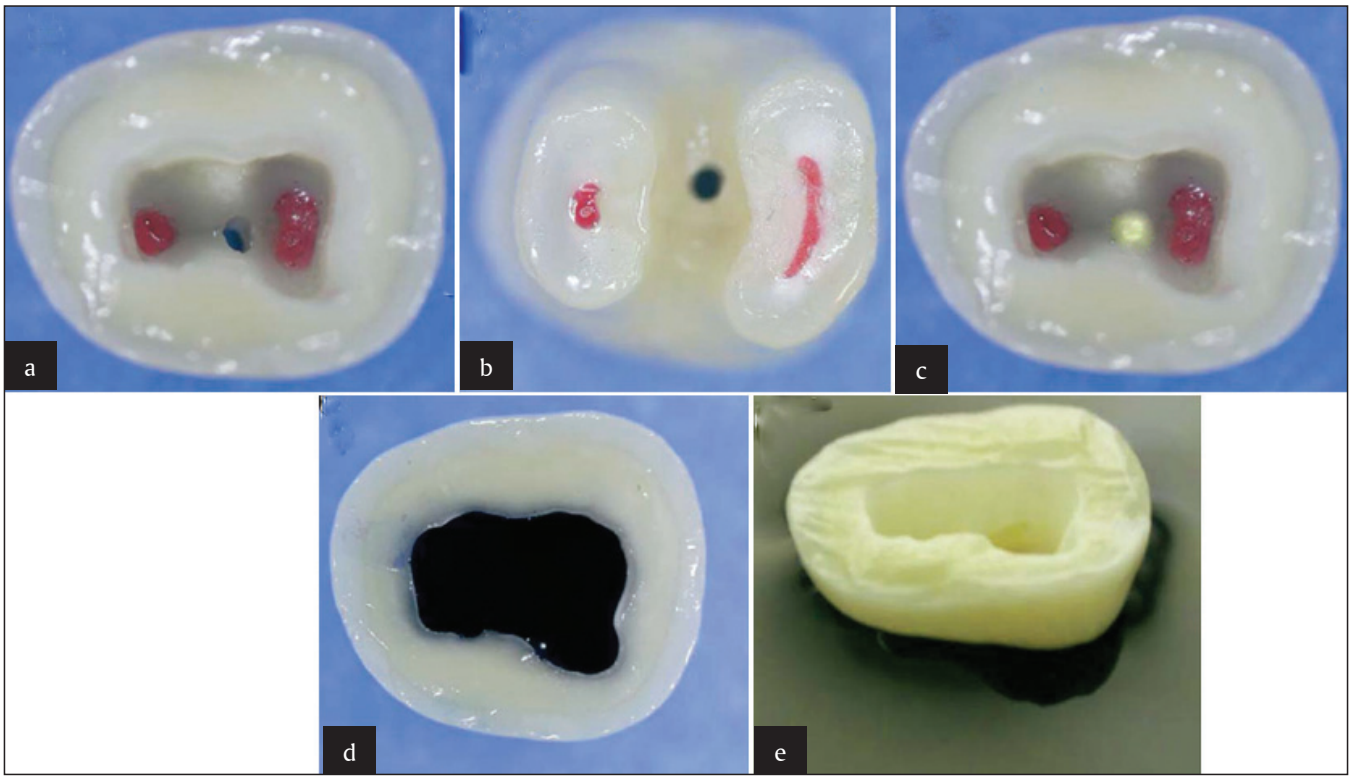

Figure 1: (a) Pulpal floor view of prepared perforation (b) furcal view of perforation. (c) Repair of perforation. Dye challenge from the (d) orthograde direction. (e) Tooth after submegence in dye for retrograde challenge.

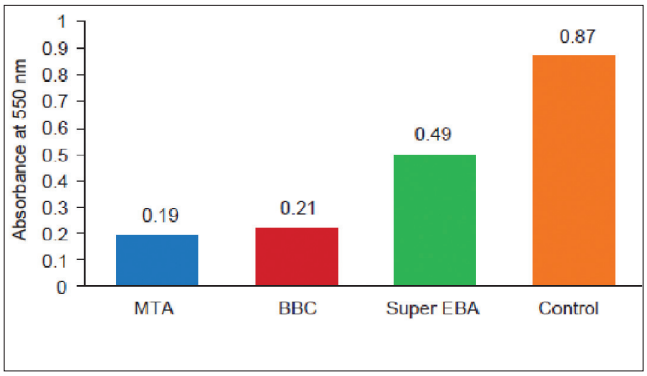

Figure 2: Absorbance values of experimental groups

bioactive bone cement (Group II) but lower than the control group. The dye absorbance values of MTA (0.19 ± 0.07379) and bioactive bone cement (0.21 ± 0.07379) were the lowest among the four groups and were not significantly different from each other.

DISCUSSION

The current study evaluated the sealing ability of bioactive bone cement, MTA and Super EBA as furcation repair materials in mandibular molars using a dye extraction leakage model. According to Ohtsuki and Kokubo *et al*. the property of bioactivity can be induced in a biomaterial through the incorporation of silanol (Si-OH) groups and calcium (Ca^{2+}) salts. The Ca^{2+} salts start triggering the formation of HA while silane coupling agent (MPS) may provide a Si-OH group due to hydrolysis of alkoxysilane after exposure to stimulated body fluid/phosphate buffered saline.[7]

Table 2: Comparison between individual groups

Comparison	Comparison difference	*Q* value	*P* value
MTA versus BBC	0.02000	0.7947	0.5620
MTA versus Super EBA	−0.3000	11.921	0.0669
MTA versus Control	−0.6800	27.020	0.0669
BBC versus Super EBA	−0.2800	11.126	0.0669
BBC versus Control	−0.6600	26.226	0.0669
Super EBA versus Control	−0.3800	15.100	0.0669

MTA: Mineral trioxide aggregate, BBC: Bioactive bone cement, EBA: Ethoxybenzoic acid

In the current study, we used MTA to modify the bone cement as it had a proven track record with favorable properties of a repair material, was easily obtainable, is chemically stable and could readily provide calcium salts when dissolved in tissue fluids[8] thus conferring bioactivity. The reason for addition of silane coupling agent (MPS) was three fold;[9] (1) it accelerates apatite formation through heterogeneous nucleation, (2) maintains mechanical properties of the bone cement by reducing amount of calcium salts incorporation, (3) increases compressive strength by bringing about chemical bonding between filler calcium salts and polymerized methylmethacrylate.

In this study, dye extraction methodology was employed which according to Camps J and Pashley[10] gave similar results to the fluid-filtration technique as both are based on quantitative measurements of liquid passage within interfaces. Moreover, it is easy to perform and does not require an elaborate equipment setup.[11] The cytotoxicity tests done on bone cement by various authors[12,13] demonstrated that fibroblast cells were completely unaffected by bone cement and its cytotoxic effect was comparable to MTA. The biocompatibility of MTA has been assured by several *in vivo* animal studies.[14] Thus, the combination of the two completely biocompatible materials can be considered to produce minimal cytotoxicity and maximum biocompatibility.

In the current study, the poorest seal was provided by Super EBA. Inability to properly condense it due to paste like consistency could have led to voids formation.[15] It was also moisture sensitive.[16] MTA provided a superior seal in comparison to Super EBA. Previously done fluid filtration study by Camps and Pashley Also recorded similar results.[17] The reason for this may be attributed to the formation of a layer of HA on the surface of MTA that forms a chemical bond with dentin which becomes stronger with time. Bioactive bone cement was able to provide a better seal in comparison to Super EBA. Previous studies by Holt and Dumsha, indicated that seal provided by Super EBA and bone cement were comparable to each other.[18] The better seal achieved with bioactive bone cement may be due to its modification with MTA which conferred bioactivity and improved bonding with the tooth structure.

Bioactive bone cement showed equally effective sealing ability as MTA. The basis for this is that on exposure to simulated tissue fluid, it gets covered with layer of apatite crystals which nucleate and grow, filling the microscopic space between bone cement and the dentinal wall.[10] Being osteoinductive in nature, it acts as a medium for crystal growth and nucleation.

It was also observed that the cement had excellent handling properties, could be easily manipulated in to a dough form and placed to adapt to the furcation area readily. Modification with MTA did not affect the setting time and was able to set even in simulated moist conditions. The exothermic reaction of polymethylmethacrylate (PMMA) bone cement during its setting does not seem to have any negative effects, even in the large quantities needed during total hip arthroplasty. The amount required in endodontics is much less, which would produce a smaller exothermic reaction and a much reduced amount of free monomer.[12] Controlled placement of repair material in furcation defects using a barrier was not considered and may prove to be a drawback in the study.

From the current study, it is obvious that bioactive bone cement was able to seal furcation perforation, similar to MTA and superior than Super EBA. Furthermore, it had better handling properties, shorter setting time and was unaffected in presence of moisture, thus, having no tendency to wash out.

The current study is perhaps the first attempt to adapt the use of bioactive bone cement in to the field of endodontics where the search for the ideal repair material is a continuing process. Further research is needed to assess the bioactive property of the bone cement used in this study, especially on the longevity of calcium release that can occur from the cement, long-term stability in relation to body tissues and tissue fluids after placement should be clarified in order to achieve successful application.

CONCLUSION

Within the limitations of this study, it can be concluded that the seal provided by bioactive bone cement in furcation repair was superior to Super EBA and comparable to MTA. It had better working properties, which could overcome potential disadvantages faced with MTA. Bioactive bone cement thus seems to be an excellent and promising material for furcation repair and also has potential to be used as a repair material in many other clinical scenarios such as apical surgery, resorptive defects, or root end filling.

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