

British Journal of Medicine & Medical Research 19(11): 1-8, 2017; Article no.BJMMR.14050 ISSN: 2231-0614, NLM ID: 101570965



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Comparison of Acidic versus Alkaline Environment for Furcation Perforation Repair among Calcium Silicate Based Materials: An *in vitro* Comparative Study

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Authors' contributions

This work was carried out in collaboration between all authors. Authors BT and BD designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author MC guided us throughout the design of the study. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BJMMR/2017/14050 <u>Editor(s)</u>: (1) Manuele Mancini, Department of Restorative Dentistry and Endodontics, University of Rome "Tor Vergata," Rome, Italy. (2) Oswin Grollmuss, Head of Department of Pediatric and Adult Resuscitation Congenital Heart of Centre Chirurgical Marie Lannelongue, University Paris XI, France. <u>Reviewers</u>: (1) Clovis Monteiro Bramante, Universidade de São Paulo, Brazil. (2) Ibrahim Hamouda, Umm Al Qura University, Saudi Arabia. (3) Masataka Yoshikawa, Osaka Dental University, Japan. (4) Nikoloudaki Georgia, University of Athens, Greece. Complete Peer review History: <u>http://www.sciencedomain.org/review-history/17928</u>

Original Research Article

Received 16th September 2014 Accepted 26th December 2016 Published 22nd February 2017

ABSTRACT

Aim: The aim of this study was to compare the effect of acidic and alkaline environment on the dislodgement resistance of White Pro Root Mineral Trioxide Aggregate and Biodentine when used as furcation perforation repair materials.

Methodology: Eighty, human, mandibular molars were used. Perforations were made in the furcation of each molar and enlarged to #3 Peeso drills. After perforation repair, according to the two materials respectively, specimens of each material were randomly divided into 4 groups (n = 10) according to storage media and time: group A: phosphate-buffered saline (PBS) (pH = 7.4) for 4 days, group B: acetic acid (pH = 5.4) for 4 days, group C: PBS for 34 days, and group D: acetic

acid (pH = 5.4) for 4 days followed by exposure to PBS for 30 days. Dislodgment resistance was then measured using a universal testing machine. **Results:** Biodentine resisted dislodgement more efficiently than Pro Root MTA in PBS (p<0.05). The dislodgement resistance of Biodentine and Pro Root MTA was reduced after exposure to acidic pH, but there was significant reduction of Pro Root MTA in comparison with Biodentine. **Conclusion:** An acidic environment affects the bond strength of calcium silicate based materials. Biodentine has better dislodgement resistance than White Pro Root Mineral Trioxide Aggregate (MTA).

Keywords: Acidity; biodentine; furcation perforation; mineral trioxide aggregate; pH; universal testing machine.

1. INTRODUCTION

Preserving the coalition of the natural dentition is mandatory for full function and natural aesthetics [1].

Endodontic therapy plays a vital role in achieving this goal. Technical problems do occur occasionally during endodontic treatment, one of which is perforation of the root canal wall during mechanical debridement. This can significantly impact the long-term prognosis of the tooth [2].

Successful management of furcation perforations poses a challenge for a clinician [3]. The perforation can result from iatrogenic causes, caries, or resorption [3]. It is advisable to repair the perforation as soon as it is identified, since any delay will allow the bacterial ingress leading to complicated endodontic-periodontal lesion [4]. Infected or inflamed tissue may have a normal pH of 7.4 or an acidic pH as low as 5.0. Acidic pH may inhibit setting reaction, affect adhesion, or increase solubility of materials. If infection or inflammation persists, erosion of filling materials can occur in acidic environment generated by bacteria or inflammation [5]. Also, exposure of root end filling material to an alkaline environment after pretreatment with calcium hydroxide might affect its properties [6]. Therefore, sealing ability of material may be directly or indirectly affected by environmental conditions and pH of medium [5].

Over the years various root end filling materials such as gold-foil, silver posts, amalgam (with and without bonding agent), zinc oxide eugenol, glass ionomer cements, mineral trioxide aggregate have been used as retrograde fillings [7]. In the recent decades, mineral trioxide aggregate (MTA) has shown promising results when used as repair material of lateral root walls or furcation perforations, root-end filling, apical plug, and root canal filling [8,9]. It consists of a fine powder of tricalcium silicate, dicalcium silicate, tricalcium aluminate, tetracalcium aluminoferrite, and bismuth oxide. During clinical application as rootend and perforation filling material or as an apical plug in necrotic teeth with open apices, MTA may be exposed to an acidic environment because of the presence of periradicular inflammation [10,11]. pH change of host tissues because of the presence of pre-existing disease may affect physical and chemical properties of material [12,13]. It has been reported that hardness [14], diametric tensile strength [11], push-out bond strength [13], and sealing ability of MTA [15] decrease after placement in acidic environment. As MTA has been shown to be suitable material, other calcium silicate-based materials have been developed recently to improve MTA drawbacks such as prolonged setting time, difficult handling, high cost, and potential tooth discoloration.

Recently, a new bioactive calcium silicate cement, Biodentine (Septodont, St. Maur-des-Fossés, France), was launched on the dental market denoted as a dentine substitute. Biodentine consists of a powder in a capsule and liquid in a pipette. The powder mainly contains tri-calcium and dicalcium silicate, the principal component of Portland cement and MTA, as well as calcium carbonate. Zirconium dioxide serves as contrast medium. The liquid consists of calcium chloride in an aqueous solution with an admixture of polycarboxylate. The powder is mixed with the liquid in a capsule in a triturator for 30 seconds. During the setting of the cement calcium hydroxide is formed. The consistency of Biodentine reminds of that of phosphate cement. Comparable to MTA, Biodentine can be used for the treatment of root perforations or of the pulp floor, internal and external re- sorption, apical plug formation, root-end filling, pulp capping and pulpotomy, but also for temporary sealing of cavities, and cervical fillings [16,17].

Nonsurgical intra canal placement of CSM materials is the preferred method of furcal perforation repair [3]. Since the teeth are unavoidably subjected to masticatory forces; dislodgement resistance of the repair material plays an important role on the time of placement of permanent coronal restoration [18].

The aim of periradicular surgery is to remove the cause of disease and provide a conducive environment for surgical wound healing. Placement of a root-end filling is one of the key steps in managing root end [19]. However, periapical lesions affect pH with consequent alteration of bond strength of root end filling materials.

There are very few studies evaluating the dislodgement resistance of different repair materials sealing the furcal perforations. The aim of the current study was to evaluate the effect of altered pH on push-out bond strength of White Pro Root MTA and Biodentine.

2. METHODOLOGY

This study was conducted in the Department of Conservative Dentistry and Endodontics, Sharad Pawar Dental College & Hospital, Datta Meghe Institute of Medical Sciences, Maharashtra, India during the months of April – May 2014.

2.1 Selection of Sample and Specimen Preparation

This study was approved by the ethical review board of Datta Meghe Institute of Medical Sciences (Deemed University). Eighty, human mandibular, first molars; with completely formed roots, minimal or no caries, no cracks, and no fused roots were selected. Mandibular molar's extracted due to periodontal reasons were used for study purpose. The selection was based on the degree of separation so as the furcal are to be visible enough. Immediately after extraction the roots of each tooth was cleaned of soft tissues or bone remnants with periodontal curettes, disinfected with Sodium Hypochlorite (NaOCI) 3% (Vishal) for 10 minutes and stored in normal saline solution until usage. The teeth were stored in normal saline to prevent desiccation of the teeth due to loss of moisture.

Molars were amputated 3 mm apical to the furcation at a direction perpendicular to the long axis of the tooth using a water-cooled, diamond disc (Brasseler Dental Products, Savannah,

USA). Similarly the crowns were also shortened to a level 3 mm coronal to the cement enamel junction using the water-cooled, diamond disc (Brasseler Dental Products, Savannah, USA). Endodontic access cavity was made in each molar, using a #4 diamond round bur (SS White, Germany), followed by safe-end bur (Mani, India).

2.2 Preparation of Furcation Cavities for the Push-out Assay

Perforations were created using size ½-round bur on a long neck (LN Bur; Maillefer, Dentsply, Switzerland) between the canal orifices in an apical direction down to the external surface of the tooth at the furcation area. The perforation was then enlarged using Peeso drills (Mani, Tochigi-ken, Japan) from size 1 to 3, corresponding to diameter of 1.1 mm, in a direction parallel to the long axis of the tooth.

The specimens were randomly divided into 2 equal groups according to the repair material, consisting of forty teeth each (n=40): White Pro Root MTA and BD. The specimens in each group were further randomly divided into 4 equal subgroups, consisting of ten teeth each (n = 10)according to the storage media and time to which the repair materials were exposed as follows: group A: specimens were placed for 4 days in contact with PBS at pH of 7.4, group B: specimens were placed for 4 days in contact with an acidic acetic acid solution buffered to pH of 5.4, group C: specimens were placed for 34 days in contact with PBS, and group D: specimens were placed in contact with acetic acid for 4 days followed by 30 days in contact with PBS. The specimens were inserted in PBS-moistened floral foam to simulate as much as possible clinical conditions.

The repair procedure was performed under 14 x magnification using surgical microscope (Opmi-Pico; Karl Zeiss, Jena, Germany) in the Phosphate Buffered Solution (PBS) media. The materials were mixed according to the manufacturers' instructions. They were applied into the perforation in increments using the micro apical placement system (MAP; Products Dentaires SA. Vevey, Switzerland) and condensed using endodontic pluggers. A cotton pellet soaked with the assigned solution for each subgroup was placed in the pulp chamber over the repair material, and the specimens were inserted in the floral foam moistened with the same respective solution. The specimens were stored at 37° C and 100° relative humidity. The soaked cotton was changed every 4 days.

2.3 Assessment of Dislodgment Resistance

A plunger tip (1.0 mm diameter) was positioned over the test material. The load was applied over the test material. The load was applied in a coronal-apical direction. Loading was performed on a universal testing machine Instron (Model LRX-Plus; Lloyd Instruments Ltd, Fareham, UK) with a load cell of 5 kN and at a cross head speed of 0.5 mm min⁻¹ until material dislocation occurred. A load x time curve was plotted during the test, using a real-time software program. The load at failure (recorded in Newton) was divided by the area of the bonded interface to express the bond strength in MPa.

The calculation of the push-out bond strength (MPa), as an indicator for dislodgment resistance, was done using the following equation:

Push-out bond strength (MPa) = Force to dislodgement (N) / Adhesive surface area (mm^2)

The adhesion area of the root canal material was calculated using the formula: area = 2π r h, where π = the constant 3.14, r = radius of the perforation cross-section, and h = height of the perforation [3].

Adhesive surface area (mm²) = $2\pi rh$,

2.4 Data Presentation and Analysis

The main outcome variable in the present study was the push-out bond strength (MPa). One-way ANOVA was used to assess the effect of each endodontic cement on push out bond strength for the groups with same storage time (4 or 34 days).

3. RESULTS

All the specimens had measurable push-out values, and no premature failure occurred. There were significant differences among the materials (p<0.05). Biodentine specimens had better push-out bond strength values (p<0.05) on exposure to PBS and acetic acid solution after 4 days and 34 days, respectively than that of White Pro Root MTA.

Dislodgment Resistance of calcium silicate based materials in various clinical situations have been depicted in Table 1.

4. DISCUSSION

Irrespective of location or aetiology, a perforation, hampers the prognosis of endodontic therapy.

The mechanical or pathological communication between root canal system and external tooth surface should be sealed with a biocompatible material as soon as possible [3].

The endodontic material used to seal the external surface of the teeth must be able to prevent leakage and remain in place under various dislodging forces, such as functional pressures or the application of other restorative materials [20].

Russian biologist Metchnikoff reported first evidence of acidic pH inside phagocytes in 1893. Later, Jensen and Bainton (1973) demonstrated that pH of a phagosome was reduced to approximately 6.5 within 3-4 min after initiation of phagocytosis. Also pH of pus aspirated from periapical tissues has been confirmed as acidic (6.68±0.324) [12]. Under certain clinical applications calcium silicate-based materials are used for the repair of root and furcation perforations, root-end fillings, and apical plugs. They are often placed in an environment where inflammation may be present and surface of unset material exposed to low pH. This altered pH may affect its physical and chemical properties [19,21].

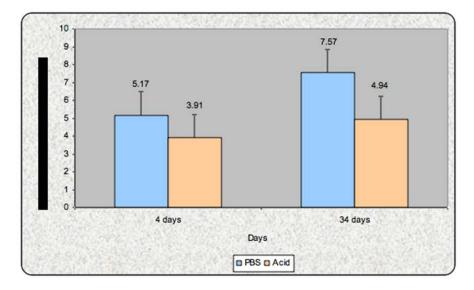
Normal tissue pH is 7.4 but it can be affected by certain clinical conditions. Tronstad et al. [22] showed pH in the range of 6.4-7 in the pulp, dentin, cementum, and periodontal ligament of vital or necrotic pulp teeth. As calcium hydroxide is preferred intracanal medicament, after its placement pH values of the most inner part of circumpulpal dentin change to pH range 11.1-12.2. It might be beneficial to provide pretreatment with calcium hydroxide in necrotic open apices or root perforations before application of biomaterials [23,24]. There are conflicting results regarding the effect of calcium hydroxide dressing on sealing ability of various biomaterials [25,26]. It has been suggested that residual calcium hydroxide might interfere with material adaptation to the root canal walls or chemically interact with them.

Materials	Group 1		Group 2		Group 3		Group 4	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Pro Root WMTA	5.17	±1.33	3.91	±1.28	7.57	±1.30	4.94	±1.27
Biodentine	8.62	±2.25	4.66	±0.94	16.92	±3.30	10.32	±0.91

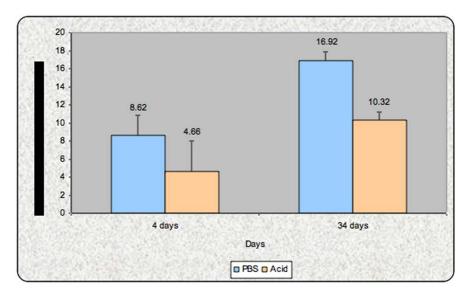
Table 1. Means of white pro root MTA and biodentine

Group 1: Specimens placed for 4 days in contact with PBS at pH of 7.4 Group 2: Specimens placed for 4 days in contact with an acetic acid solution buffered to pH of 5.4 Group 3: Specimens placed for 34 days in contact with PBS

Group 4: Specimens placed in contact with acetic acid for 4 days followed by 30 days in contact with PBS



Graph 1. Represents means, standard deviation and statistical analysis of push out bond strength of white pro root MTA



Graph 2. Represents means, standard deviation and statistical analysis of push out bond strength of biodentine

In the present study, push-out bond strength of Biodentine and White Pro Root MTA was evaluated and compared after exposure to acidic and alkaline pH. For acidic environment, acetic acid buffered at pH 6.4 was used. For alkaline environment phosphate buffer saline solution at pH 7.4 was used.

In this study, the samples were placed for four days in PBS (pH 7.4), four days in acetic acid (pH 4.4), thirty four days in PBS (pH 7.4) and four days in acetic acid (pH 4.4) followed by thirty days in PBS (ph 7.4) during setting and thereafter; in order to simulate physiologic, pathologic, and healing conditions respectively to evaluate the push-out bond strengths of Biodentine and White Pro Root MTA.

The present results pointed out a significantly different performance among the tested materials at both situations (four days exposure to acidic environment alone and four days exposure to acidic environment followed by thirty days in alkaline environment). Biodentine produced higher bond strength values than White Pro Root MTA (p < 0.05).

Prominent biomineralization ability of Biodentine can most likely be attributed to the formation of tags, and may be the cause of its superior dislodgement resistance herein demonstrated. Han and Okiji [27] showed that uptake of calcium and silicon ion into dentin, leading to the formation of tag-like structures in Biodentine, was higher than in MTA, and may have a role in the overall adaptation to root dentin. Moreover, the dissimilar particle sizes of MTA and Biodentine may affect their penetration into the dentinal tubules, with consequences to displacement resistance.

Atmeh et al. [28] demonstrated that Ca-Si cements facilitate the permeation of Ca and OH ions into the dentine, due to their caustic effect. It can be assumed that the improved ability to release remineralising Ca and OH ions by Biodentine is responsible for the improved formation of apatite at the interface [29,30], and for micromechanical anchorage [31].

Biodentine showed better push-out bond strength values at four days in acidic medium when compared to those of White Pro Root MTA (p < 0.05).

Erosion on the surface of cubic crystals, absence of needle-shaped crystal formations, increased porosity and decreased portlandite formation (the main product of MTA hydration, which is calcium hydroxide in crystal form) were observed after MTA placement in an environment with a pH value of 5. The absence of needle-like crystals in the acidic pH might be attributed to higher surface area and the crystals' rapid dissolution in the acidic environment. Since porosity results in the progression of solubility, the increased MTA solubility in the present study might be attributed to increased porosity and changes in MTA crystalline structure after exposure to an acidic environment [32]. This is a possible explanation as to the decrease push-out bond strength in an acidic environment of White Pro Root MTA.

Even though in acidic environment the push-out bond strength of both calcium silicate materials have decreased, Biodentine still shows an increased resistance to dislodgement than White Pro Root MTA. This might be explained on the basis of calcium chloride present in the liquid. The addition of calcium chloride is intended to reduce the setting time of Portland cement and improve its physicochemical properties as explained by Poplai et al. [33]. A possible explanation behind calcium chloride enhancing the physical properties are that calcium chloride penetrates the pores of cements, strongly accelerating the hydration of silicates and leading to their faster crystallization and reducing the setting time.

Similarly in these both situations (four days exposure to acidic environment alone; and four days exposure to acidic environment followed by thirty days in alkaline environment), Biodentine shows higher push-out bond strength values. To simulate healing process, depicted in the present study as exposure to four days in acidic environment followed by thirty days in alkaline environment, the push-out bond strength of Biodentine (16.92 MPa) and of MTA (10.32 MPa). MTA showed inferior bond strength in all environmental conditions, which is in agreement with Shokouhinejad et al. [13] It is possible that pH inhibits setting reaction, affects adhesion, or increases solubility of calcium silicate-based materials eventually affecting mechanical properties of material including compressive strength.

In the current study, Biodentine showed greater bond strength in in all conditions than MTA, apart from the fact that its push-out bond strength was altered in certain conditions. Bond strength of MTA was most likely affected by the alkaline pH of dentin [15]. Biodentine demonstrated superior bond strength in alkaline medium than in acidic conditions. In the presence of tissue fluid, the hydration of calcium silicate–based materials results in the formation of hydroxyapatite crystals and the development of a hybrid layer between dentin and calcium silicate–based materials. The development of hydroxyapatite crystals and consequently a hybrid layer at the calcium silicate–based materials–dentin interfacial gap are likely to be disturbed in an acidic environment [19].

5. CONCLUSION

Under the conditions of this study, it can be concluded that effect of acidity decreases the bond strength of calcium silicate based materials. Biodentine seems more appropriate for use when exposed to an acidic environment compared with White Pro Root MTA.

CONSENT

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history: The peer review history for this paper can be accessed here: http://sciencedomain.org/review-history/17928