

Comparison of the Sealing Ability of MTA-Angelus, Biodentine and CEM Cement in the Repair of Large Furcal Perforations-A Bacterial Leakage Study

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ABSTRACT

Introduction: Materials such as MTA-Angelus, Biodentine, and CEM Cement have been recommended for the repair of large furcal perforations. Due to larger surface area of such perforations, evaluating the sealing ability of the aforementioned repair materials is important for their clinical selection.

Aim: To compare the sealing ability of MTA-Angelus, Biodentine, and CEM cement when used as repair materials for large furcal perforations.

Materials and Methods: Sixty-five extracted human molar teeth were used for the study. Samples were randomly divided into groups 1, 2, and 3 with 20 samples in each (n=20). Five samples were used as controls. Furcal perforations of standardised diameter (2 mm) were prepared in samples of groups 1, 2, and 3 and repaired with MTA-Angelus, Biodentine and CEM Cement, respectively. A bacterial leakage model was used for each sample to study the sealing ability of these repair materials over

an experimental period of 50 days. A culture of *Enterococcus faecalis* and sterile Brain Heart Infusion (BHI) broth were placed into the upper and lower chambers of the model, respectively. Any turbidity of the BHI broth indicated bacterial leakage through the repaired perforations. The day wise number of samples with bacterial leakage and the percentage of these samples during every five-day interval of the experimental period were noted and the results were statistically analysed using Chi-Square and Log Rank (Mantel-Cox) tests. The level of significance was set at p-value less than 0.05.

Results: There was no significant difference in the bacterial leakage among the three groups at any 5-day interval of the experimental period (p>0.05).

Conclusion: Biodentine and CEM cement with better handling properties could be used as alternatives to MTA-Angelus while repairing furcal perforations.

Keywords: Calcium-enriched mixture, Furcation involvement, Sealability

INTRODUCTION

Perforations of the pulp chamber and the root canal system adversely affect the prognosis of endodontic treatment. Ingle JL reported root canal perforation as the second most common cause of endodontic failures as it accounted to 9.6% of all unsuccessful cases [1]. Furcal perforation is an artificial communication between the pulp chamber and the supporting structures of the tooth through the floor of the pulp chamber. It can occur due to a large carious lesion, pathological resorption, or iatrogenic mishap during endodontic treatment. Furcal perforations can lead to periradicular break-down with eventual loss of gingival attachment and bone [2,3].

A furcal perforation can be repaired using a non-surgical or a surgical approach. However, the latter is less preferred due to difficulty in obtaining accessibility for repair. Moreover, it often leads to loss of attachment, pocket formation, and periodontal furcation involvement [4]. Therefore, a minimally invasive nonsurgical approach through the coronal access is recommended to repair a furcal perforation [4].

Factors that affect the prognosis of perforation repair includes the level and the location and size of the perforation, the time delay before the perforation repair and the material used for sealing the perforation [5]. A gamut of materials including zinc-oxide eugenol cements (IRM and Super-EBA), glass ionomers, resin-glass ionomer hybrids, and composite resins have been suggested for repair of furcal perforations. Ideally, a perforation repair material should be biocompatible, well-sealing, non-resorbable, radiopaque, and bacteriostatic [6]. Since large furcal perforations act as a bottomless pit, the extrusion of a repair material is unavoidable during perforation repair through the coronal access [7]. Moreover, large

furcal perforations are difficult to completely seal off with a repair material due to their size and extent. Therefore, a material used for the repair of a large furcal perforation should be biocompatible with shorter setting time and good sealing ability [7,8].

Various calcium silicate-based materials have been recommended for the repair of furcal perforations due to their sealing ability, biocompatibility, regenerative capability, and antibacterial property [9-12]. One such commonly used material is Mineral Trioxide Aggregate (MTA) which is shown to be biocompatible as a repair material. It is available in commercial forms such as ProRoot MTA (Dentsply, Switzerland) and MTA-Angelus (Angelus, Londrina, Brazil) [13]. However, MTA-Angelus is preferred for the repair of furcal perforations due to shorter setting time and better handling properties [14,15].

Biodentine™ (Septodont, Saint Maur des Fossés, France) is another calcium silicate-based repair material available for furcal perforations. It has good handling, biological, mechanical and physical properties [16]. Similarly, a novel calcium silicate-based material called Calcium-Enriched Mixture (CEM) cement (BioniqueDent, Iran) has also been suggested for the repair of furcal perforations due to its biocompatibility and regenerative properties [17].

Even though there have been studies comparing MTA with other calcium silicate-based materials, there is sparse data comparing the sealing ability of MTA-Angelus, Biodentine, and CEM cement for repairing perforations in permanent teeth [18]. Hence, this study was conducted to compare the sealing ability of MTA-Angelus, Biodentine, and CEM cement when used as repair materials for furcal perforations, using dual-chamber bacterial leakage model.

MATERIALS AND METHODS

This in-vitro study was carried out in KLE VK Institute of Dental Sciences, Belagavi, Karnataka, India. The ethical clearance for performing this study was obtained from Institute Ethics Committee (Registration no. ECR/211/Inst/KA/2013) and the duration of this study was two years from March 2013-March 2015. Sixty-five extracted human maxillary and mandibular permanent molar teeth were collected and placed in 0.5% sodium hypochlorite (NaOCl) solution for three days to remove organic debris and disinfect them.

Preparation of the Samples

Access openings were prepared on these teeth using an endo access bur (Dentsply, Switzerland) and apical few millimeters of the roots were sectioned to facilitate the placement of teeth into the bacterial leakage model of the study. The exposed root ends were sealed with a cyanoacrylate resin (Fevikwik, Pidilite, India) and all the surfaces of teeth except the furcation area were painted with two coats of nail varnish (Nail Trend, Fiabila, India) to obtain the study samples.

The samples were randomly divided into groups 1, 2, and 3 with 20 samples (n=20) in each. Remaining 5 samples were used as controls to check the effectiveness and reliability of the bacterial leakage model of the study. In samples from groups 1, 2, and 3, furcal perforations of standardised diameter (2 mm) and centered between the roots were prepared by holding teeth in hand and perforating the chamber floor from the external surface using a no. 4 long shank carbide round bur (SS White Burs, New Jersey, USA). However, no furcal perforations were prepared in the controls.

The furcal perforations in groups 1, 2, and 3 were repaired with MTA-Angelus, Biodentine, and CEM cement, respectively. These repair materials were mixed according to their manufacturer's instructions and placed into the furcal perforations. A moist cotton pellet was placed within the pulp chambers and samples were covered with moist gauze and left for 72 hours at 37°C in an incubator to allow the setting of repair materials.

Bacterial Leakage Model

A bacterial leakage model, based on a design as suggested by Barthel CR et al., was used to study the sealing ability of the aforementioned repair materials [19]. It was designed for each sample by using a 5 mL centrifuge tube (HiMedia laboratories, Mumbai, India) and a 15 mL glass vial with a screw cap (Riviera Glass Private Limited, Mumbai, India) which served as the upper and lower chambers of the model, respectively. The hinged cap of the centrifuge tube was cut-off 10 mm from the tip. This tube served as the upper chamber of the model. It was snugly fitted around the crown of the sample in order to project the internal side of the repaired furcation towards the upper chamber. The lower chamber of the model was designed by making a hole through the centre of the screw cap of glass vial using a round bur. The upper chamber with the sample was fitted into the hole of screw cap in order to project the external side of the repaired furcation towards the lower chamber [Table/Fig-1]. The interface of sample and centrifuge tube was double-sealed with cyanoacrylate resin followed by heat-moistened sticky wax (DPI Model Cement, DPI, India) to complete the upper chamber. The interface of upper chamber and screw cap was also sealed in a similar way. These components of the model were sterilised overnight using an ethylene oxide gas steriliser (3M™ Steri-Vac™ Sterilizer, St. Paul, MN, USA). Sterile Brain Heart Infusion (BHI) broth (HiMedia laboratories, India) was placed into the glass vial to complete the lower chamber. The screw cap with sealed upper chamber was fitted onto the glass vial to complete the bacterial leakage model.



[Table/Fig-1]: Bacterial leakage model used in the study.

Bacterial Leakage and its Monitoring

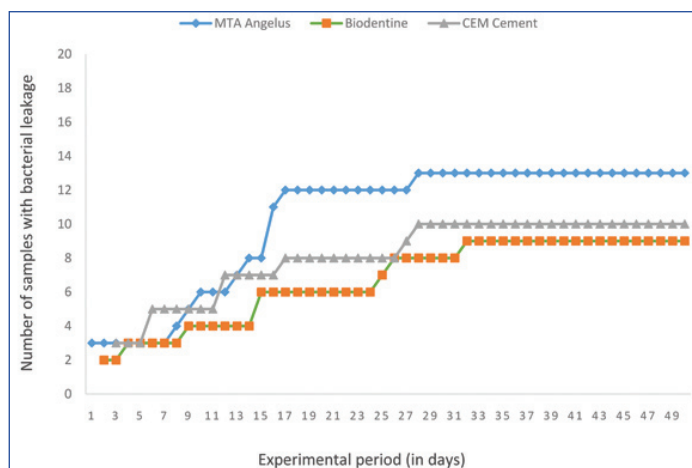
This procedure was carried out under aseptic conditions in a laminar flow chamber (Yorco Sales, New Delhi, India) for an experimental period of up to 50 days. Using a sterile micropipette, 0.1 mL overnight broth culture of *Enterococcus faecalis* was placed into the upper chamber of the model. The model was placed in an incubator maintained at 37°C. Fresh overnight culture of the organism was placed into the upper chamber every alternate day to ensure the viability of *E. faecalis*. The sterile BHI broth in the lower chamber of the model was monitored daily for any turbidity which denoted bacterial leakage into the broth due to penetration of *E. faecalis* through the repaired furcal perforation and lack of sealing ability of the repair material. In case of any turbidity, a 10 µL aliquot of the turbid broth was taken and streaked onto a plate containing BHI agar and incubated for 24 hours. This was done in order to observe the colony morphology of *E. faecalis* and rule out any contamination by other microorganisms. The day wise number of samples with bacterial leakage and the percentage of these samples during every 5-day interval of the experimental period of 50 days were noted.

STATISTICAL ANALYSIS

The results were statistically analysed using Chi-Square and Log Rank (Mantel-Cox) tests. Statistical analysis was performed using Statistical Package for the Social Sciences (SPSS, software version 17.0, IBM, Chicago, IL, USA). The level of significance was set at p-value less than 0.05.

RESULTS

None of the control samples showed any bacterial leakage throughout the experimental period which indicated the effectiveness and reliability of the model employed in our study. The day wise number of samples with bacterial leakage and the percentage of these samples during every 5-day interval of the experimental period of 50 days are shown in [Table/Fig-2,3]. During the first 5 days, all the 3 groups had equal amount of bacterial leakage, following which the leakage observed was greater in group 1 (MTA-Angelus). By the end of 50th day, 65% (13/20) samples in group 1 (MTA-Angelus) had bacterial leakage which started between 1st and 28th day. Similarly, 45% (9/20) samples in group 2 (Biodentine) had bacterial leakage which started between 2nd and 32nd day. Whereas, 50% (10/20) samples in group 3 (CEM cement) showed bacterial leakage which started between 3rd and 27th day. However, statistical analysis of the samples with bacterial leakage showed no significant differences among the three groups at any 5-day interval of the experimental period (p>0.05).



[Table/Fig-2]: Day wise number of samples with bacterial leakage in the experimental period of 50 days.

Number of 5-Day Interval	Group 1 (MTA-Angelus) (n=20)	Group 2 (Biodentine) (n=20)	Group 3 (CEM Cement) (n=20)	p-value*
1 st (1 st -5 th day)	15%	15%	15%	1
2 nd (6 th -10 th day)	30%	20%	25%	0.766
3 rd (11 th -15 th day)	40%	30%	35%	0.802
4 th (16 th -20 th day)	60%	30%	40%	0.149
5 th (21 st -25 th day)	60%	35%	40%	0.243
6 th (26 th -30 th day)	65%	40%	50%	0.281
7 th (31 st -35 th day)	65%	45%	50%	0.418
8 th (36 th -40 th day)	65%	45%	50%	0.418
9 th (41 st -45 th day)	65%	45%	50%	0.418
10 th (46 th -50 th day)	65%	45%	50%	0.418

[Table/Fig-3]: Intergroup comparison of percentage (%) of samples with bacterial leakage during every 5-day interval of the experimental period of 50 days.

*Chi-Square test, p-value <0.05 is statistically significant. N: Number of samples

DISCUSSION

The main prognostic factor in the management of a furcal perforation is the time lapse between its occurrence and repair [20]. Therefore, immediate repair of a furcal perforation is important for endodontic success. However, profuse bleeding from the perforation site would limit the clinician from immediate sealing with conventional restorative materials. On the other hand, a repaired furcal perforation itself could lead to inflammation in the adjacent area due to poor sealing ability and/or cytotoxicity of the repair material [6,8,21]. Therefore, calcium silicate-based materials such as MTA-Angelus, Biodentine, and CEM cement could be advantageous as repair materials. The sealing ability of these materials becomes important when they are chosen for the repair of a large furcal perforation due to the need for sealing a larger surface area [8]. Therefore, it is clinically relevant to evaluate and compare sealing ability of the aforementioned materials.

In this study, bacterial leakage model with anaerobic bacteria *E. faecalis* was employed because the detection of bacteria would clinically reflect better upon the sealing ability of repair materials as bacterial leakage is the cause for all periradicular pathosis and most bacteria causing endodontic infections are anaerobes [22]. Bacterial leakage model is shown to provide biologically significant data that are clinically relevant when compared with other methods [23-25].

In the present study, although MTA-Angelus showed relatively more bacterial leakage when compared with Biodentine and CEM cement, but statistically there is no significant difference in the sealing ability of these materials when employed for the repair of a large furcal perforation.

MTA is reported to exhibit favorable sealing property due to cementogenic activity as it releases calcium ions which interact with phosphate groups in the surrounding tissue fluid to form

hydroxyapatite on its surface [26]. However, in a study conducted by Brito-junior M et al., MTA-Angelus exhibited 70% bacterial leakage within 20 days [27]. The findings of our study are in accordance with this study. It was hypothesised that the shorter setting time of MTA-Angelus may prevent it from having good wetting and adaptation to the walls of a defect and could lead to higher bacterial leakage [28].

Biodentine is recommended as a perforation repair material because it has good mechanical strength and is biocompatible and bioactive [29]. It has better handling properties and shorter setting time compared to MTA [30]. In this study, though there was no statistical difference, Biodentine showed relatively lesser bacterial leakage compared to MTA-Angelus. This could be attributed to the ability of Biodentine to form and precipitate hydroxyapatite [31]. It is also capable of inter-tubular diffusion and formation of mineral tags of hydration products leading to hybrid zone formation with dentine [32]. According to a study by Guner MB et al., Biodentine showed significantly higher push-out bond strength than MTA when used as root perforation repair materials [33]. Moreover, Biodentine shows better interlocking with dentine compared to MTA because of its smaller particle size and uniform components [33].

CEM cement is another novel material recommended for repair of perforations. It has a setting time of less than one hour with more flow and less film thickness compared to MTA [34]. Previous studies have shown that the sealing property of CEM cement is not significantly different from MTA [35,36]. The findings from this study are in agreement with those studies. However, CEM cement showed relatively lesser bacterial leakage compared to MTA-Angelus though there was no statistical difference. CEM cement could be expected to display better sealing due to many reasons. It has higher content of endogenous phosphates leading to more hydroxyapatite precipitation, particularly in an aqueous environment, when compared with MTA [37]. Furthermore, on hydration CEM cement shows slight expansion leading to better marginal adaptation. Additionally, CEM cement has good flow, less film thickness, and better handling and chemical properties which could also contribute towards better sealing [37,38].

LIMITATION

The main limitation with bacterial leakage study is that, it does not simulate the conditions of the oral cavity and require observation for long periods of time. As the results of this in-vitro study, may not demonstrate the full clinical potential of the materials used to seal perforation defects, we suggest future in-vivo researches to evaluate the sealing ability of the tested materials.

CONCLUSION

Within the limitations of the present study, it could be concluded that there is no significant difference in the sealing ability of MTA-Angelus, Biodentine and CEM cement when employed as a repair material for large furcal perforations. Hence, the newer biomaterials, Biodentine and CEM cement with better handling properties could be used as alternatives to MTA-Angelus while repairing furcal perforations.

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