

Evaluation of Mineral Trioxide Aggregate and Biodentine as Direct Pulp-capping Agents in Human Teeth: An Ex-Vivo Study

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ABSTRACT

Introduction: Biodentine (Tricalcium Silicate based) is a modern bioactive cement that is comparable to the broadly utilized mineral trioxide aggregate (MTA). It has dentin-like mechanical properties, which can be considered appropriate material for diseased dentin-pulp complex recovery such as direct pulp capping. The present study was to compare the reaction of the pulp-dentin complex in human teeth after direct pulp capping with use of MTA and Biodentine.

Objective: To assess the histomorphologic response of human dental pulps on direct pulp capping with MTA and Biodentine.

Methods: Pulp of 34 maxillary permanent intact human third molars planned for extraction were mechanically exposed with sterile rotary round bur and allotted to one of the two exploratory groups, MTA or Biodentine. After four weeks, the teeth were extracted, stained with hematoxylin-eosin, and categorized by employing a preset histologic scoring framework. The histopathologic evaluations scored data were recorded and statically analyzed using SPSS where Mann-Whitney U test was used and $p < 0.05$ was considered statistically significant.

Results: No inflammatory pulp reaction seen in 34 (100%) teeth whereas 19 (55.9 %) showed complete dentinal bridge arrangement. Layers of well-arranged odontoblast and odontoblast-like cells were found to make tubular dentin beneath the osteodentin. Inferential statistics using showed insignificance between the MTA and Biodentine test groups amid the perception period, showing similar outcomes.

Conclusions: Biodentine had comparative adequacy in the clinical setting and may be considered an appropriate alternative to MTA as the pulp-capping agent.

Keywords: Biodentine, direct pulp capping, histology, MTA, pulpal reaction.

INTRODUCTION

Conservative pulp treatments are performed to preserve the coronal and radicular pulp tissue in a viable condition. The treatments range from vital pulp treatments (VPT) including direct pulp capping (DPC), and partial and total

pulpotomy to the more intrusive pulpectomy and root canal treatment.^{1,2,5} Numerous studies support calcium hydroxide as the benchmark for VPT. However, with it, there are limitations like the presence of tunnels defects, extensive dentin formation obliterating the pulp chamber, high solubility in oral fluids, and lack of adhesion and degradation after acid etching.^{3,4}

MTA is a bioactive, biocompatible, antibacterial material with desired properties and high sealing capacity. It has become a standard material for this reason. The shortcomings of MTA like long setting time and difficult handling issues,⁶⁻⁹ have led to the development of Biodentine, a modern tricalcium silicate-based therapeutic cement with dentin-like mechanical properties that positively impact vital pulp cells and induces tertiary dentin formation.^{10,11}

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The present study aims to evaluate the clinical and histologic responses of the pulp-dentin complex after direct capping with the MTA and Biodentine in human teeth after four weeks.

METHODS

This is an ex-vivo study conducted from March 2016 to March 2018. The study obtained ethical clearance from the institutional review committee, BPKIHS (31/071/072-IERB), and was supported by Nepal-India Corpus Fund in BPKIHS. Patients were educated about the rationale of the study, clinical strategies, and conceivable complications. This was followed by informed signed consent forms. All procedures performed in this study followed the ethical standards of BPKIHS and the 1964 Helsinki announcement and its afterward amendments or comparable ethical standards.

The sample size was calculated by taking Alicja Nowicka et al.,² 2013 study into consideration. The minimal sample size calculated was 30 with 15 in each group (10% additional people were recruited on the note of lost to follow up. Thus, the total sample was 34 with power = 90% and $p < 0.5$ with 17 people in each group.

Clinical Procedure

The study procedure followed Alicja Nowicka, et al.² protocol. Patients aged 21 years and above with upper third molars destined for extraction due to their position causing trauma to the cheek were included in the study. The tooth was examined clinically and radiographically to ensure the absence of proximal caries and periapical lesions. Patients with features of irreversible pulpitis or caries involving the inner third of the dentin verified radiographically were excluded from the study.

The operative procedure was carried out in upper third molars meeting inclusion criteria. All procedures were performed by a single operator in the Department of Conservative Dentistry and Endodontics.² Endo-Ice (Coltene/Whaledent Inc, Mahwah, NJ) test was used to confirm the vitality of the teeth. Before the operative procedure, the tooth was sanitized and cleaned with 0.2% chlorhexidine solution. After local anesthesia (2% Lidocaine, 1:80,000 Adrenaline; Septodont, Saint Maur des Fosses, France), the rubber dam isolation was done.

The occlusal cavity was prepared using an air turbine with sterile 1.3 mm diamond points (SF 31, Mani Inc, Japan) at high speed under water/air coolant. Once the exposure was made, hemostasis was established with a sterile cotton pellet soaked in saline solution.

In group I patients, white mineral trioxide aggregate (Angelus Industria Deproductos Odontologicos S/A, Brazil) and in group II patients, Biodentine (Septodont, Saint Maur des Fosses, France) was placed in the occlusal floor as direct pulp capping agent with subsequent Type IX glass ionomer cement (Fuji IX, GC Corporation, Japan) intermediate restoration.

The patients were asked about the presence or absence of postoperative sensitivity or any other discomfort on the 2nd day and after a week after the procedure was completed.

Teeth from both groups were extracted after four weeks. The extraction was performed under local anesthesia by an Oral Surgeon in the Department of Oral and Maxillofacial Surgery, College of Dental Surgery.²

Histologic Examination

Once extracted, the apical third of all roots were sectioned in 5 mm to facilitate fixation in 10% buffered formalin solution for 72 hours. The teeth were then decalcified in 10% nitric acid for 3-4 days, prepared according to normal histologic techniques, and embedded in paraffin wax. Six-micrometer-thick sections were cut with a microtome parallel to the main vertical axis of the tooth. The sections were then mounted on glass slides, and stained with hematoxylin-eosin.²

The histomorphologic assessments were performed using modified criteria set by Faraco et al.²⁴ and Medina et al.²⁵

Continuity of the dentinal bridge

- 1 = complete dentin bridge formation
- 2 = partial/incomplete dentin bridge formation extending to more than one-half of the exposure site but not completely closing the exposure site
- 3 = initial dentin bridge formation extending to not more than one-half of the exposure site
- 4 = no dentin bridge formation

Morphology of the dentinal bridge

- 1 = dentin or dentin associated with irregular hard tissue
- 2 = only irregular hard tissue deposition
- 3 = only a thin layer of hard tissue deposition
- 4 = no hard tissue deposition

The thickness of the dentinal bridge

- 1 = >0.25 millimeter (mm)
- 2 = 0.1–0.25 mm
- 3 = <0.1 mm
- 4 = partial or absent bridge

Type of pulp inflammation

- 1 = no inflammation
- 2 = chronic inflammation
- 3 = acute and chronic inflammation
- 4 = acute inflammation

The intensity of pulp inflammation

- 1 = absent or very few inflammatory cells
- 2 = mild, defined as an average of <10 inflammatory cells
- 3 = moderate, defined as an average of 10–25 inflammatory cells
- 4 = severe, defined as an average >25 inflammatory cells

Extensivity of pulp inflammation

- 1 = absent
- 2 = mild, defined as inflammatory cells only next to dentin bridge or area of pulp exposure
- 3 = moderate, defined as inflammatory cells observed in part of coronal pulp (in one-third or more of the coronal pulp or the mid pulp)
- 4 = severe, defined as all coronal pulp is infiltrated or necrotic

The sections were blindly evaluated by an experienced and calibrated oral pathologist in the Department of Oral Pathology using a Labomed research microscope

(Labomed, India).² The amount of hard tissue formation at the interface of the capping material (continuity, morphology, and thickness), and pulp inflammation (type, intensity, and extension), were decided and scored from 1–4, with 1 speaking to the most desired result and 4 speaking to the least desired result as per the altered criteria. The thickness of the dentinal bridge was measured at the thickest, most slender, and midmost point regions of the continuous dentin bridge. The mean of the 3 values was calculated.

Statistical Analysis

The histopathologic evaluations were scored, recorded and statistically analyzed using the Mann-Whitney U test where $p < 0.05$ was considered statistically significant.

RESULTS

There were 34 patients selected within the study separated into two groups of which 11 (32.35%) patients (5 with MTA and 6 with Biodentine) complained of spontaneous minor pain after the treatment within 24 hours of surgery. They were prescribed Ibuprofen 400 mg twice a day for two days for the relief of pain. Other patients 23 (76.64%) reported no specific side effects during the test period. All teeth were sensitive to cold and electric pulp tester suggestive of vital pulp before extraction. In addition, no periapical pathologies were observed by radiography before the clinical procedure and extraction.

Histologic assessment of teeth revealed that both materials were well endured by the pulp tissue. Results of all the specimens in the MTA and Biodentine are provided in Table 1. Within the Biodentine test group, the pulp reactions were comparable to those perceived within the MTA group (Table 1). The injured site observed the dentin bridge directly underneath the capping materials with both materials. Total dentin bridge arrangement was seen in 10 (29.4%) teeth in the MTA group and 9 (26.45%) teeth in the Biodentine group in overall sample. In most samples from both the groups, dentin was related to an irregular hard tissue, but in some samples the reparative tissue appeared mixed with cell inclusions, 5.88% ($n=2$) of MTA group and 2.94% ($n=1$) of Biodentine group (Table 1). The average thicknesses of the hard-tissue dentin bridge in the Biodentine and MTA groups were 185.36 micrometers (μm) and 193.31 μm , respectively.

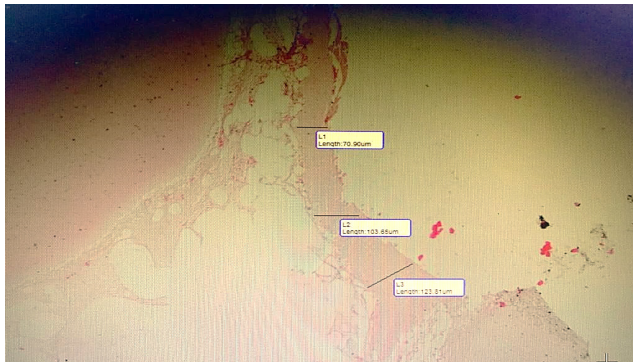


Figure 1. Shows dentinal bridge formation with MTA.

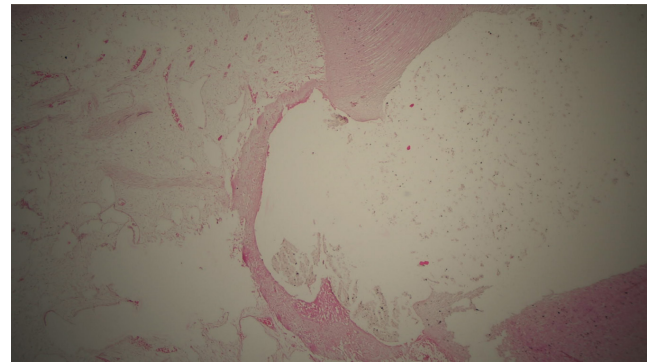


Figure 2. Shows dentinal bridge formation with Biodentine.

Table 1. Summary of different categories of histologic features^{24,25} of MTA and Biodentine according to the scores.

Experimental Groups (n=34) →	MTA (Group I; n=17)				Biodentine (Group II; n=17)			
Histologic Criteria (Scores) →	1	2	3	4	1	2	3	4
Histologic Criteria of the hard tissue evaluation								
Continuity of the dentinal bridge	10	7			9	8		
Morphology of the dentinal bridge	8	7	2		8	8	1	
Thickness of the dentinal bridge		16	1			14	3	
Histologic Criteria of Pulp Inflammation								
Type of pulp inflammation	17				17			
Intensity of pulp inflammation	15	2			14	3		
Extension of pulp inflammation		17				17		

There was no evidence of abscess, or necrosis underneath the dentinal bridge (Table 1). In regard to inflammation, there were no or few inflammatory cells seen with no dilated blood vessels in the majority of pulp specimens.

Furthermore, regarding the histologic evaluation criteria, there was no statistical significant difference between the reactions of teeth to MTA compared with Biodentine as a direct pulp-capping agent ($p > 0.05$) using the Mann-Whitney U Test.

DISCUSSION

This study presents direct pulp capping done by Biodentine with MTA in sound human third molars where a light microscopic investigation is done for comparing comprehensive information on the bridge and pulpal inflammation with the comparative result of pulp capping materials used. The results of this study appear that intentional pulp defects treated with both calcium silicate based material are naturally free from inflammation and ended up with compact, dentin-like hard tissue bridges.

Dentin formation can be a questionable issue as it can be a sign of irritation or healing.^{12,26} In this study, the

arrangement of the dentinal bridge was seen as a positive response to stimulation and a sign of healing.²⁶ The release of transforming growth factor- β 1 early by the pulp cells may have resulted in reparative dentin synthesis, by both the calcium silicate based materials.^{10,11} Alike MTA-based materials, Laurent et al.¹⁰ suggested that materials physicochemical properties might boost the mineralization process as particles of Biodentine were captured within the recently shaped foci, and mineralization showed up as osteodentin. Biodentine has both calcium and silicon ions which are its major component and this might have fortified cell multiplication and differentiation.³ Various examinations have detailed the effective application of MTA in pulp capping.^{1,3,6,14-20,26} The present study is in understanding with other studies noticing the formation of dentin bridge arrangement. In attendance with this study, dentin bridge arrangement was seen in all pulps capped with MTA as formerly observed (Table 1).^{1-3,6}

Hengameh Bakhtiar et al. did a study that evaluated the capacity of Biodentine, MTA, and Theracal to induce pulp healing after partial pulpotomy in human teeth.²⁰ Considering our study that appreciated dentin bridge in

four weeks, the investigators confirmed the arrangement of a dentin bridge at the injury site after eight weeks. The cells displayed an odontoblastic phenotype in all assessed materials. These reparative structures that were formed by both calcium silicate based materials were homogenous and in coherence with primary dentin. Comparable to our study, dentin tubules could be seen, and the cells secreting this structure showed odontoblastic characteristics.²⁰

Biodentine was tested in cell culture by Zanini et al.¹¹ This study assessed these new materials potential in the differentiation of odontoblast-like cells and biomineralization. In support of our study, research illustrating the expression of several genes confirmed the differentiation of OD-21 cells into odontoblasts during the period of cell culture and biomineralization suggestive of Biodentine's capacity to stimulate dentin-forming cells. Additionally, in a rat model study carried out by Tran et al; the investigators demonstrated the formation of a dentin bridge in the injury site after 30 days depicting results comparable to our study.¹³

Numerous authors have suggested, that pulp response after direct capping is associated with bacterial microleakage.^{3,8,23} Microbes can compromise the pulpal healing when treated with capping materials.⁶ Researches show aggravation of inflammation by bacteria results in poor dentin bridge formation despite of the material used for pulp treatment.²¹⁻²⁴ The findings have implied that pulpal survival after an oral exposure is not so much a function of an agent's potential bioactivity but its capacity to protect the pulp from bacterial exposures.^{4,6} Henceforth, averting bacterial leakage into cavity preparations is a meaningful intent in treatment planning and bestows to the longevity of cavity restorations.²³ In the current study,

all the preparations were surface sealed with type IX glass ionomer cement (GC Corp, Japan) to avert microbial leakage for a period of four weeks. The researchers evaluating the performance and safety of Biodentine²⁵⁻³⁰ found that Biodentine can be used as a posterior restoration material for up to 6 months after direct pulp capping though our study did not use Biodentine alone. It was covered with GIC as it has desired properties as a temporary restoration for adult teeth.³¹

In this study, teeth were intact with no ongoing inflammation. Direct pulp cap has various uses and with studies supporting its use in symptomatic pulpal exposure, these materials have broadened their uses.^{5,19} Therefore, within the limitation of this study conducted in non-inflamed teeth, further histologic studies can be carried out to prove its acceptability in inflamed teeth.

CONCLUSION

From the results of the study, it can be implied that Biodentine induced the formation of a reparative dentin bridge at the pulp exposure site in-vivo. Given its good bioactivities and handling characteristics, it can be used in a similar manner as an alternative to MTA in a clinical setting.

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Conflict of Interest: None

INAPD

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