



## Histological comparison of three calcium silicate materials in the Pulpotomy of rat molars

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### ABSTRACT

**Introduction:** The importance of preservation of pulp tissue prolongs the vitality of the tooth. The introduction of calcium silicate cements has proven to increase the success rate of the treatment, but the increased cost of the material is a disadvantage. Thus the development of a new material which has a superior clinical effect and is safe and economical is of utmost importance.

**Aim:** The purpose of this research is to histologically evaluate a novel calcium silicate cement and correlate it to Biodentine and Mineral trioxide aggregate (MTA) as pulpotomy agents in a rat model.

**Subjects and methods:** A total of nine Wistar albino rats were divided into three groups equally. Pulpotomies were conducted on the caries free mandibular molars using the three calcium silicate cements- new calcium silicate cement, Biodentine, and MTA under general anaesthesia. Glass ionomer cement that had been resin-modified was used to seal access cavities. At 7, 14, and 28 days following surgery, pulpotomized teeth in both groups were examined histologically under a light microscope (10x).

**Results:** Over the course of the experimented time periods, the inflammatory cell response gradually decreased in all three groups.

**Conclusions:** The new calcium silicate cement has been shown to have a less inflammatory reaction. This new calcium silicate cement can be used as a cost-effective alternative pulpotomy agent in comparison to the commercially available cements.

**Keywords:** *cost, calcium, molars, alternative*

## INTRODUCTION

The tissue present in the tooth known as the dental pulp is a connective tissue surrounded by mineralized dentin and enamel which plays a role in maintaining the vitality of the tooth, triggering immune responses, sensitivity and has also been found to stimulate repair and regeneration(1). It consists of a network of cells which are significant in producing reactions against pathogens during tissue injury or infection.(2,3). Any inflammation visualized in the dental pulp is called pulpitis. Based on the extent of damage, pulpitis can be classified into reversible and irreversible. This can be treated by vital and non-vital pulp therapy.

The main aim of performing pulp therapy in primary dentition is to retain the tooth in the oral cavity until the eruption of the succedaneous tooth to avoid infection, swelling and pain which ultimately leads to the removal of the tooth. (4). This treatment modality preserves the arch length and maintains the proper functions of the orofacial complex(5). Pulpotomy performed in primary teeth, particularly the molars, is a conservative treatment done when there is extensive caries approximating or involving the coronal pulp(6). According to the AAPD, primary teeth devoid of any radicular pathology have a long-term clinical success rate. According to Finn in 1995, pulpotomy is the removal of the complete coronal portion of the pulp followed by the placement of a suitable medicament or dressing which would result in preserving and healing the vitality of the tooth.

After achieving ample hemostasis, the exposed portion of the pulp is covered by a suitable medicament or cement. Pulpotomy was first introduced by Buckley in 1904, where formaldehyde was used. Due to the concerns of cytotoxicity and necrosis produced by formocresol(7), other reagents were studied and Calcium silicates were discovered in 1900s and were found to have high success rates(8). Mineral Trioxide Aggregate (MTA) which Torabinejad discovered was the first calcium silicate material used in dentistry and was a modification of portland cement(9). The disadvantage of MTA is the delayed time(10) it takes to set which was soon overcome with the introduction of

Biodentine by Septodont(11–13). The drawbacks of these materials are their heavy weight on the pocket(14,15). Though new alternatives are available, they do not have a good antimicrobial effect, this can be overcome with a new calcium silicate material that has been created in our institution(16,17). The action of this material has not been studied extensively, hence the present study was intended to evaluate and compare the three calcium silicate cements as pulpotomy agents in rat teeth.

## MATERIAL AND METHOD

Nine caries free Wistar male Albino rats, 8-16 weeks old with a weight ranging from 250-300 grams were involved in this study. The study clearance was obtained from the Institutional Animal Ethical committee. The subjects were placed in a room that had a constant temperature of  $25^{\circ}\text{C} \pm 1^{\circ}\text{C}$  and a 12/12-hr light and dark cycle. They were fed rat pellet feed (Biogen Animal Health product, Bangalore) and water ad libitum.

### *Procedure*

The rats were divided into three groups with three rats in each group. Pulpotomy was carried out on the mandibular right and left molars in each rat. The pulpotomies were performed and histologic analysis was carried out at 1, 7 and 28 days in Group I (new calcium silicate material), Group II (MTA) and Group III (Biodentine).

Prior to the pulpotomy procedure, the rats were anaesthetised with 22-24 mg/kg intramuscular ketamine hydrochloride (Brand). The oral cavity was cleaned with 0.2% chlorhexidine digluconate solution for one minute. After the rats were immobilized with the anaesthetic, they were positioned dorsally and a holding device was used to maintain their mouths in an open position. A round diamond bur (001/010 BR45, Mani Inc, Japan) and an Endo safe end bur (ESE-014, SS White UK) was used for the access cavities under copious saline irrigation. The pulp chambers were irrigated with sterile saline solution once the access was opened, and they were then gently dried with sterile paper points. Each silicate cement was prepared as directed by

the manufacturer. Then, resin-modified glass ionomer cement was used to fill the coronal cavities of the teeth. Rats were sacrificed with an overdose of intramuscular Ketamine 1, 7 and 28 days postoperatively.

### ***Histological procedures***

After the sacrifice, the pulpotomized teeth and surrounding soft tissue were placed in individual containers containing formalin. These samples were then sent to a lab for decalcification. All the treated teeth were prepared according to standard histological techniques, stained with H and E, and blindly evaluated under a light microscope (Leica BM E educational microscope, Leica Microsystems, Germany) at x4 and x10 calibration and scored by an oral pathologist.

## **RESULTS**

The surgical procedure was well tolerated by the experimental animals, with no evident adverse effects occurring during an observation period of 1–28 days.

Photomicrographs showing the histopathology of experimentally induced pulpitis in rats in

Group-I (New Material), Group-II (MTA) and Group-III (Biodentine) at Day 1, Day 7 and Day 28 were stained with Haematoxylin & Eosin.

The histopathological evaluation demonstrated the characteristics of inflammation after pulpotomy in all the 3 groups depicted by infiltration of inflammatory cells in the pulp cavity and the adjacent areas. The inflammatory cells were almost even in all the groups on Day 1. After the onset of inflammation, on Day 7 the inflammatory cells were increased in Group-I (New Material) when compared to Group-II (MTA) and Group-III (Biodentine). As the inflammation subsides gradually by Day 28, Group-I (New Material) revealed comparatively fewer inflammatory cells when compared to Group-II (MTA) and Group-III (Biodentine).

## **DISCUSSION**

The failure for pulpotomy can be due to incorrectly diagnosing the subclinical inflamed pulp(18). The pulp repair mechanism is

important to analyze the regenerative pulpal mechanism that occurs after applying a pulp capping agent and observe if the responses produced result in pulpal necrosis (19). The pulpal responses were studied for 24 hours, 7 days and 30 days after the conduction of the pulpotomies as inflammation is an early response (20). The method used to evaluate the pulpal response was by using animal models due to ethical issues as healthy primary molars cannot be extracted immediately after pulpotomy to evaluate the pulpal response to various materials (21). Dammuske in 2010 (22) stated that rat models could be used to test capping materials as they emit similar responses to that of humans and the mechanisms of dental pulp repair could be studied both at a cellular and molecular level(23).

The best method to assess pulp capping agents used in pulpotomies is to assess the pulpal response to the material in question(24). Clinical and radiographic methods are not precise in diagnosing and studying the pulpal responses(25,26). Histological assessments must be made to analyze the true state of the pulp. Even though the periods of observation were short, this study shed light on the cellular changes that might lead to the pulp repair after the application of the pulpotomy agents.

## **CONCLUSION**

The radicular pulp of new calcium silicate teeth resulted in better histological features and less inflammatory reaction after 28 days. This suggests that it can be used as a low-cost alternative to the other commercially available calcium silicate cements available in the market. Further studies should be conducted to confirm the histological success of the new calcium silicate cement after pulpotomy of primary teeth.

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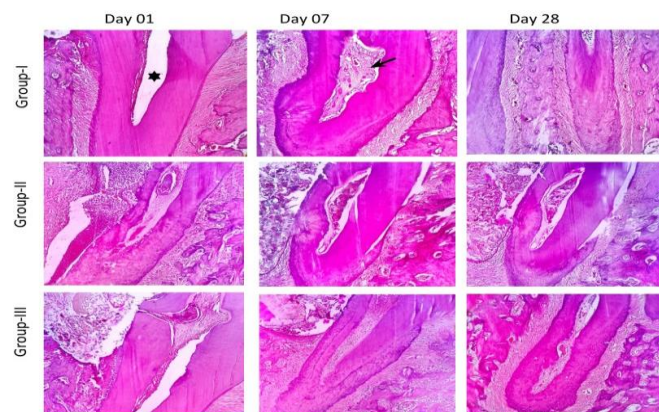
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**FIG 1** Access cavity was prepared using a round diamond bur (001/010 BR45, Mani Inc, Japan) and an Endo safe end bur (ESE-014, SS White UK)



**FIG 2** Placement of pulpotomy agent



**FIG 3** Photomicrographs showing the histopathology of experimentally induced pulpitis in rats in Group-I (New Material), Group-II (MTA) and Group-III (Biodentine) at Day 1, Day 7 and Day 28 stained with Haematoxylin & Eosin.