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DOI: 10.23804/ejpd.2017.18.03.03

Microleakage of MTA in primary molar pulpotomies

ABSTRACT

Aim *The main goal of this study was to compare the coronal microleakage of MTA and IRM®, using the dye penetration technique in primary molars.*

Materials and methods *Study design: 160 extracted primary molars (caries-free or not) were used and randomly distributed among four groups of study (n=30) and four groups of controls, two positive (n=10) and two negative (n =10). Class II type cavities were created in 140 molar crowns and 120 of those were randomly restored with IRM or MTA. A dye penetration technique was used to evaluate the coronal microleakage. One group of each restorative material and one of each positive and negative control were kept submerged in a methylene blue solution for 24 hours, and the other groups and controls were kept in the solution for 28 days. The penetration of the colouring solution was examined under the microscope in two different locations, the coronal axial wall and the proximal cavity.*

Results *The samples of the positive control group showed full penetration of blue methylene and the samples of the negative control group showed no penetration at all. MTA showed lower filtration than IRM; the results were statistically significant (p<005) in both axial and proximal cavity. Both IRM and MTA groups did not revealed significant differences at 24 hours and 28 days. Statistics: The SPSS 13.0 software was used for the statistical analysis of the collected data. The study results were analysed using an independent sample t-test and the one-way Anova test.*

Conclusions *The results of this study show that, when used in primary molar pulpotomies MTA showed in fluid filtration tests results significantly lower than obturations with IRM.*

Keywords *Coronal microleakage; Dye Penetration; MTA; Primary teet; Pulpotomy.*

Introduction

Primary tooth pulpotomies are an important part of the daily routine of clinicians who work with children and paediatric dentists. This procedure may fail if the pulpotomy agent is not the most adequate, if the intermediate restorative material is not effective and if the final obturation is not the correct one.

Over the years, various materials have been used in pulpotomies, such as formocresol [Eidelman et al.., 2001], calcium hydroxide, glutaraldehyde ferric sulphate, zinc oxide, freeze-dried bone, bone morphogenetic protein, osteogenic protein, electrosurgery, laser, cement calcium enriched CEM® [Mehrdad et al.., 2013].

Formocresol [Walker et al.., 2013] is the most widely used material due to its good clinical results. However, its biocompatibility is still unclear as several studies show that it is a potentially carcinogenic material [Sun et al., 2013]. The use of formocresol as a pulpotomy agent has been questioned for many years. Currently, concern has been expressed about its immune sensitisation, toxicity [Menezes et al., 2009] and mutagenic and carcinogenic potential in humans [Casas et al., 2005]. Therefore, it is necessary to find a non-carcinogenic material with good physical and chemical properties in order to protect both patients and professionals who use these materials on a daily basis. In this regard, Mineral Trioxide Aggregate, or MTA, in pulpotomies of primary teeth [Foley et al., 2013] has been used as an alternative to formocresol. Several studies performed using MTA as a pulpotomy agent have confirmed its biocompatibility [Souza Costa et al., 2008] and its ability to promote calcified tissue formation [Cardoso-Silva et al., 2011]; they also showed the formation of a thin layer [Zhu et al., 2000] due to the adhesion of odontoblast cells to MTA, which induces the formation of a dentin bridge [Maroto et al., 2005]. Both White and Grey MTA seem to induce dentin bridge formation, but the Grey MTA showed a stronger effect [Maroto et al., 2006]. MTA also appears to be effective in the treatment of pulpitis caused by bacterial infection [Cannon et al., 2008]. It is also non-mutagenic [Kettering et al., 1995]. Recent studies have confirmed that the chemical composition and characteristics of MTA and Portland cement are similar [Sakai et al., 2009]; however, Portland cement does not contain bismuth and is not sold in small sterilised packages.

The appropriate temporary filling material is a key factor and can determine the success or failure of pulpotomy. Such material seals the tooth, preventing fluids, microorganisms, and other debris from entering in the root canal space. MTA reveals very good sealing properties in dry and blood-contaminated environments [Maturo et al., 2009]. MTA has also been shown to have the necessary compressive strength [Parirokh et al., 2010]; its compressive strength at 21 days is approximately 70 Mpa, the same as Intermediate Restorative Material (IRM)®, a zinc oxide-eugenol based material [Torabinejad et al., 1999].

It is crucial to use a provisional material with good sealing ability. Due to the good biological performance, in primary teeth pulpotomies with MTA, it might be more practical to fill the entire cavity because of MTA's mechanical stability. A stainless steel crown will then be placed over most primary teeth [Hutcheson et al., 2012]. Ideally [Seale, 2002], the stainless steel crown should be placed immediately after the pulpotomy, which is often not possible due to the prolonged operative time and the patient's unwillingness to undergo both procedures on the same visit to the dentist.

If cavities are successfully sealed, just like when using MTA [Barrieshi-Nusair and Hammad, 2005], this may prevent marginal leakage caused by the dissolution of the zinc phosphate cement used to cement the crowns. In pulpotomies with formocresol, the remaining cavity is often filled with IRM® (which is the commercial form of zinc oxide-eugenol with higher mechanical strength) before making a stainless steel crown.

The aim of this study was to observe the characteristics of marginal seal of MTA, when used as a temporary filling material after pulpotomies, and compare it with the most widely used material, IRM®. A simulation of cavity preparation for pulpotomy was conducted with one compromised axial wall; this means that in this study we used a cavity larger than those used in most of the reported studies.

Materials and methods

In this study, 160 extracted primary molars, with at least 2/3 of root length still present and restorable, were collected due to orthodontic reasons or caries in the Paediatric Dentistry Department of the Odontological Hospital of the Dental Medicine Faculty of the University of Barcelona. From these 160 teeth, 52 (33%) were lower second molars, 42 (26%) lower first molars, 34 (21%) upper first molars and 32 (20%) upper second molars.

The extracted primary molars, were randomly distributed among four groups of study (n=30) and four control groups (n=10). Two study groups were filled with MTA ProRoot and one was left to soak in the dye for 24 hours (MTA ProRoot 24 hours) and the other group for 28 days (MTA ProRoot 28 days); the same for the IRM, (IRM 24 hours) and (IRM 28 days). The four control groups were divided into two positive and two negative controls. No filler was added to the chamber of the two positive control groups. One positive control group was left to soak in the dye for 24 hours and the other for 28 days. No crown prep work was made in the teeth of the two negative control groups; one group was left to soak in the dye for 24 hours and the other one for 28 days (Table 1).

	24 hours	28 days	Total
IRM	30	30	60
MTA Pro Root	30	30	60
Positive Control	10	10	20
Negative Control	10	10	20
Total	80	80	160

TABLE 1 Study groups. Samples were divided in 4 study groups: 1 group was treated with IRM, 1 with MTA, 1 positive control and 1 negative control. Each group was then divided in 2 subgroups; one group was left to soak for 24 hours and the other one for 28 days.

After extraction, the molars were placed in a sterile saline solution (sodium chloride 0.9%, Laboratories ERN, Barcelona, Spain) to prevent dehydration. Organic debris were removed with curettes or excavators (Excavator round 1.5, LM dental, Parainen, Finland) and a toothbrush. The same operator made the class II cavities on 140 molars; the remaining 20 were randomly removed to the negative control group.

The opening of the tooth was performed using a high-speed dental turbine with a cooling unit. All carious tissue, if present, was removed with a round steel bur (Dentsply, Maillefer, Ballaigues, Switzerland). A cavity was made in the centre of the clinical crown to reach the pulp chamber. The cavity was then extended using a cylindrical bur (Dentsply, Maillefer), to reach one of the proximal walls, simulating a caries affecting 1/3 of the center of the tooth's occlusal surface and one of the proximal surfaces. The basic rules of operation were respected:

a) remove all unsupported enamel;
b) set margins in an area of self-cleaning or easy cleaning;
c) prepare tooth for pulpotomy.

After reaching the pulp chamber, we used an Endo-Z (Dentsply, Maillefer) cutter without the active tip; this allowed us to widen the camera lens in order to see all the canal entrances, when looking at the tooth from occlusal angle.

Pink wax (Reus wax, Reus, Spain) was placed in the roots of the teeth and covered with varnish (nail polish, Vera Valenti), to keep the roots sealed and to prevent dye penetration. They were covered with three layers of varnish, except the surface access and each layer was left to dry for 24 hours.

The height of the preparation was measured as well as the buccolingual width of the coronal cavity and the width of the proximal box. For 140 molars an average depth of 5.2 mm was found, the average width was 3.6 mm and the size of the proximal box in buccolingual direction was 3.6

	Qualification criteria:
0	No penetration
1	Penetration to the enamel-dentin union
2	Penetration up to dentin, without reaching the pulp chamber
3	Penetration up to the entries of the ducts

TABLE 2 Qualification criteria. Microleakage grading criteria.

mm. The teeth were then distributed randomly between the two groups of positive control and the 4 groups of study. Of the 140 teeth, 47 (34%) were lower second molars, 37 (26%) lower first molars, 31 (22%) upper first molars and 25 (18%) upper second molars. A metal matrix 0.03 mm thick (KerrHawe, Washington DC, USA) was then placed in the port-matrix; we then adjusted the port-matrix to the size of each tooth (3073, Asa Dental).

MTA (ProRoot White, Dentsply) was prepared according to manufacturer's recommendations; the powder was mixed in a beaker with sterile water at a 3:1 ratio and mixed with a metal spatula to obtain a paste with a grainy consistency. This paste was then placed in an amalgam carrier, so we could reach the intended location, pressed against the canal with a burnisher, pressing it gently with a slightly moistened swab. Then the entire cavity was filled.

IRM was prepared according to manufacturer's recommendations (1:1 liquid to powder). When a white non-sticky paste is obtained, it means it is ready to be placed and inserted in the cavity, and then pressed with a wetted cotton swab.

One group was sealed with MTA, one was sealed with IRM, one with positive control and one with negative control and all were then inserted in a 2% aqueous solution of methylene blue (Pharmacia Gema Garrigós, Barcelona, Spain). They were kept in this solution for 24 hours and the other subgroup for 28 days. Methylene blue was used because it is known for being a sensitive indicator of microfiltration and for having a molecular size similar to microorganisms.

Teeth were cleaned and prepared for mesial-distal cuts. First, some cuvettes (for making ice) were carefully smeared with vaseline (Medical, Franco-Portuguese Laboratory, Lisboa, Portugal) to prevent the applied resin from sticking to the cuvette. The cuvette provides shape, so that samples can be cut. Then the teeth were placed in the best position for cutting, i.e. the cross-sectional area parallel to the base of the cuvette. The acrylic resin (Resin 2195) was then mixed with the catalyst and the blocks were left to rest for at least 24 hours, to enable a smooth polymerisation. When the resin hardens it forms a solid block that holds the tooth in place allowing the microtome to cut this block into sections.

A microtome (Accutom-5, Struers) was used to section the teeth; first, the blocks were removed from the cuvettes, then placed in the microtome, which must be securely fastened or clamped and tightened with key. When securely fastened, it is necessary to bring the clamp closer to the blade. The microtome was previously configured as follows: Cutting modality: tooth; Disk: 230 CA; Spin: 3000 rpm; Strength: low.

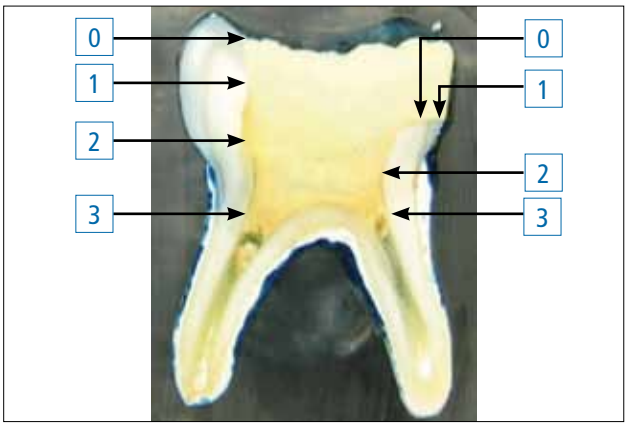


FIG. 1 Grade of leakage (measures the penetration of the dye). 0: no penetration; 1: penetration to the enamel-dentin junction; 2: penetration up to the dentinal tubules but not reaching the pulp chamber; 3: penetration up to the entrance of the canals.

The teeth were then sectioned into mesial and distal parts; in the end the resin block was partially attached to the clamp and detached at the base of the microtome plate.

An optical microscope (PhotomakRosk M400, Wild Heerbrugg, Switzerland) 80x was used to visualise the dye penetration, which was observed and recorded for the two values of each tooth, one showing the linear penetration in the axial coronal wall and the other in the proximal box. The qualification criteria were (Table 2) (Fig. 1): no penetration (0) no ink in the enamel tissue; penetration up to the enamel-dentin junction (1), penetration into the enamel but not into the dentin; penetration up to the dentinal tubules but not reaching the pulp chamber, (2) the ink reached the dentin but not the pulp chamber; penetration up to the entrance of the canals (3) dye penetration in all the tissues up to the entrance of the canals.

All data were processed using SPSS software (13.0, SPSS Inc., Chicago, USA). The study results were analysed using the t-test of independent variables and the one-way Anova test . The P-value was < 0.05 and considered statistically significant.

Results

All positive controls showed complete dye penetration, whereas negative controls showed no penetration.

IRM revealed a mean penetration for the axial wall and the proximal box of 2.35 and 2.58, respectively; the average in the axial wall and in the proximal box is 3 (Table 3) (Fig. 2). MTA showed a mean penetration of 0.15 and 0.40, respectively and the average was 0 for both axial wall and proximal box (Table 4) (Fig. 3). Significant differences were found ($p<005$) in both axial wall and proximal box.

The observed differences were not due to random chance but to the materials used.

The results of the IRM group at 24 hours and 28 days were similar. The same was observed in the MTA groups. This means that in this case the main factor is the material used, and not the time.

Discussion

One of the main goals of paediatric dentistry is to preserve primary teeth, until natural exfoliation occurs. It is thus necessary to find materials with the right physical and chemical [Fuks, 2002] properties for pulpotomy and MTA seems to be one of them. Several studies have compared different pulpotomy agents and in all of them the clinical success rates of MTA were higher [Arien et al., 2012; Sushynski et al., 2012], 97% MTA and 85% formocresol. The radiographic success rates for MTA and formocresol were 97% and 83%, respectively [Holan et al., 2005]. In another 24-month study, MTA revealed a clinical and radiographic success rate of 100%, whereas formocresol 98.6% and 86.8%, respectively [Farsi et al., 2005]. When compared with calcium hydroxide (64%) MTA showed a clinical and radiographic [Moretti et al., 2008] success rate of 100%. Also when compared with ferric sulphate, MTA revealed a clinical success rate of 94.7% and a radiographic success rate of 92.1% against 84.7% and 78.2%, respectively [Odabas et al., 2012].

This proves the need for a material with good biological properties and good sealing abilities, since many times the crowns that are placed immediately after pulp treatment (during the same visit to the dentist) not always fit well on the prepared tooth. In two studies on microfiltration using intact primary teeth, the prepared cavities were smaller, 2x2 mm [Odabas et al., 2009] and 3x3x1.5 mm [Dos Santos et al., 2014], than those prepared in this study (3.6x3.6x5.2), for intact or carious primary teeth. Most of the times, primary teeth have at least one compromised proximal wall; therefore the mechanical characteristics of

IRM					
Samples	24 hours		Samples	28 days	
	Axial coronal wall	Proximal box		Axial coronal wall	Proximal box
1	2	2	31	2	3
2	1	2	32	1	3
3	2	3	33	1	3
4	2	2	34	1	3
5	2	3	35	2	1
6	2	1	36	2	3
7	2	3	37	2	2
8	1	1	38	2	2
9	1	1	39	3	3
10	3	3	40	3	2
11	3	3	41	2	3
12	3	3	42	3	3
13	3	3	43	3	2
14	2	2	44	3	3
15	2	2	45	3	3
16	3	2	46	3	3
17	2	2	47	3	3
18	3	3	48	3	3
19	3	3	49	3	3
20	3	3	50	3	3
21	3	3	51	2	2
22	3	3	52	2	3
23	3	3	53	3	2
24	1	2	54	3	3
25	2	3	55	2	3
26	3	3	56	3	3
27	3	3	57	3	3
28	2	2	58	2	3
29	3	3	59	2	2
30	1	2	60	2	3

TABLE 3 Results for IRM penetration in 60 samples. 24-hour setting: samples 1-30 and 28-day setting: samples 31-60.

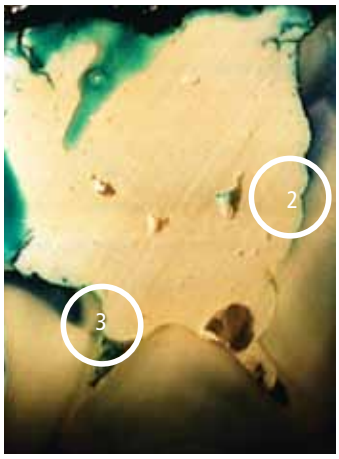


FIG. 2 Sample number 5 (IRM). Image of a molar filled with IRM. A grade 2 leakage can be observed on the axial wall and a grade 3 on the proximal box.

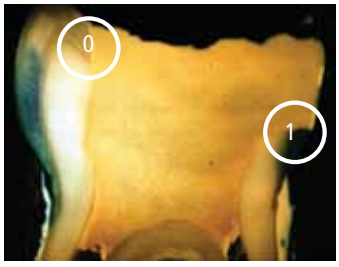


FIG. 3 Sample number 61 (MTA). Image of a molar filled with MTA. A grade 0 leakage (no leakage) is observed on the axial wall and a grade 1 leakage on the proximal box.

MTA					
Samples	24 hours		Samples	28 days	
	Axial coronal wall	Proximal box		Axial coronal wall	Proximal box
61	0	1	91	0	0
62	0	2	92	0	0
63	1	1	93	0	0
64	0	2	94	0	2
65	1	1	95	0	0
66	1	1	96	0	3
67	0	0	97	0	0
68	0	2	98	1	1
69	0	0	99	0	0
70	0	0	100	0	0
71	0	0	101	0	0
72	0	0	102	0	0
73	0	0	103	0	0
74	0	0	104	0	0
75	0	0	105	2	2
76	0	0	106	0	0
77	0	0	107	0	0
78	0	0	108	0	0
79	0	0	109	0	0
80	0	2	110	1	0
81	0	0	111	2	0
82	0	1	112	0	1
83	0	0	113	0	0
84	0	1	114	0	0
85	0	1	115	0	0
86	0	0	116	0	0
87	0	0	117	0	0
88	0	0	118	0	0
89	0	0	119	0	0
90	0	0	120	0	0

TABLE 4 Results for MTA penetration in 60 samples. 24-hour setting: samples 61-90 and 28-day setting: samples 91-120.

the filling material are of the utmost importance.

In this study we compared the characteristics of MTA, used as provisional filling material, with the most widely used material, the IRM, after a pulpotomy with formocresol or ferric sulphate.

The coronal microleakage of MTA was lower than the one observed with IRM. MTA showed a significantly higher sealing ability when compared to IRM and this trend was constant at least over a period of 28 days. MTA results revealed to be more successful, both in the axial and in the proximal wall box.

The hourly results were consistent with those obtained in other studies, although many were not specific for primary teeth, and used cavities smaller than the ones used in this study. One should note that most of the reviewed microleakage studies were performed in caries-free permanent teeth. Therefore, it is important to highlight that due to the lack of scientific papers on primary teeth and to the fact that most studies mentioned in the literature were performed using permanent teeth, the results may sometimes differ from the ones shown in this study. More complex studies using larger samples have to be developed.

Conclusion

According to the results of this study, we may conclude that MTA shows very low leakage when used as restorative material in the restoration of primary molars. MTA obturations on primary molars showed in fluid filtration tests results significantly lower than obturations with IRM material.

When used in primary molar pulpotomies MTA is a material that produces a proper seal and prevents fluid filtration, which allows it to be indicated as a substitute to formocresol in pulpotomies of primary molars without presenting any adverse results.

Acknowledgements

We would like to thank the Department of Endodontics of the University of Barcelona for letting us use all the instruments needed to complete this article.

Conflict of interest

The authors declare that there is no conflict of interest.

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