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Clinical, Radiographic and Histological Evaluation of Primary Teeth Pulpotomy Using MTA And Ferric Sulfate

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The aim of this study was to evaluate the clinical, radiographic and histological outcomes of the dentin-pulp complex from primary molars after pulpotomy with mineral trioxide aggregate (MTA) and 15.5% ferric sulfate (FS). Thirty-one primary molars were randomly allocated into MTA or FS groups. Clinical and radiographic evaluations were recorded at 3-, 6-, 12- and 18-month follow-up. Teeth at the regular exfoliation period were extracted and processed for histological analysis. Clinical and radiographic data were tested by statistical analysis ($p \leq 0.01$). Histological outcomes were analyzed descriptively. All of the treated teeth presented clinical success over the experimental periods. Both groups exhibited 100% of radiographic success at 3, 6 and 12 months. At the 18-month follow-up, one tooth from FS group presented inter-radicular radiolucency ($p > 0.01$). Histologically, the treated teeth presented pulp vitality and absence of inflammatory infiltrate into the connective tissue. Only MTA group showed hard tissue barrier surrounded by odontoblasts over the pulp stumps. Both MTA and 15.5% FS are effective for pulpotomies of primary teeth. Although MTA is considered the first-choice material, FS may be a suitable alternative when treatment cost is an issue.

Key Words: ferric sulfate, mineral trioxide aggregate, pulpotomy, deciduous tooth.

Introduction

Nowadays, pediatric endodontics focuses on keeping the pulp of primary teeth vital until their physiological resorption in order to maintain the space between teeth, prevent detrimental tongue and speech habits, preserve esthetics, and maintain chewing function (1,2).

Pulpotomy is a common therapy for asymptomatic cariously exposed pulp with no involvement of the radicular portion, thus avoiding the premature loss of the tooth (1). Successful pulpotomy procedure depends not only on the correct diagnosis of the inflamed dental pulp, but also on the selection of an effective and biocompatible medicament (1,3). The ideal capping material should be bactericidal and harmless to cells and surrounding structures, promote healing of the pulp tissue and not interfere with the physiologic root resorption (4-6). A recent systematic review found no evidence to clearly identify one superior pulpotomy material, although two medicaments seem to be preferable: mineral trioxide aggregate (MTA) and ferric sulfate (FS) (7).

MTA consists of fine hydrophilic powder, has the ability of maintaining pulp vitality and promoting repair when placed in contact with dental pulp or periradicular tissues (1,3). In root canals, MTA promotes an active deposition of mineralized tissue, with radicular stenosis or pulp canal obliteration (2,3,5). Although MTA presents high clinical, radiographic and histological success rates (2,5,6), its high cost is a remarkable inconvenient since pulpotomy is considered a low-cost technique, with unquestionable social value (7). For this reason, other therapies have been investigated in order to find a low-cost pulp capping material with success rates similar to MTA.

In some countries, FS has been considered a low-cost alternative pulp capping material, due to its high clinical and radiographic success rates (4,8-10). The reaction of FS with blood results in a ferric ion-protein complex, which mechanically blocks the capillary orifices, favoring the hemostasis (4,9).

Despite the favorable success rates (11), few studies evaluate the microscopic effects of FS on the pulp tissue (12). Thus, the purpose of the present study was to evaluate the clinical, radiographic and histological outcomes of the dentin-pulp complex from primary molars after pulpotomy with white MTA and 15.5% FS.

Material and Methods

Participants

This study was approved by the Human Research Ethics Committee of the Federal University of Alfenas (protocol #158/2011) and followed the principles outlined by the Helsinki Declaration. Participant screening was performed at two Brazilian state schools from Alfenas city through dental clinical evaluation using wood spatula under natural light. Selected children were invited to participate of a

thorough clinical and radiographic evaluation at Dental School of Federal University of Alfenas. The parents or guardians of the children received detailed information concerning the procedures involved in the study and signed informed consent forms.

The inclusion criteria for the study were: children aged between 5 and 9 years old, primary maxillary or mandibular molars compromised by deep caries cavities with vital pulp; absence of history of spontaneous pain; no clinical or radiographic evidence of fistula or abscess, absence of internal and external root resorption, inter-radicular and/or furcal bone destruction; and the possibility of proper restoration of the teeth. Exclusion criteria consisted of the presence of systemic pathology and history of allergic reaction to latex, local anesthetics, or to the constituents of the pulp dressing agents tested (5).

Clinical Procedures

Two trained and experienced pediatric dentists performed the pulpotomies with a standardized single-visit treatment protocol (5). After local anesthesia using 2% lidocaine and 1:100.000 epinephrine and rubber dam isolation, caries was removed by round carbide bur at low speed (#3, KG Sorensen, Cotia, SP, Brazil) and the opening of the pulp chamber was conducted with round burs (#1014, #1015, KG Sorensen, Cotia, SP, Brazil) at high speed, under water spray. Coronal pulp tissue was removed manually with an excavator (SS White, Dental Objects Ltda., Juiz de Fora, MG, Brazil), followed by irrigation with saline solution to clear off the debris and the placement of a dry sterile cotton pellet for 5 min to achieve bleeding control.

In the MTA group, the pulp stumps were covered with white MTA paste (MTA White, Angelus, Londrina, PR, Brazil) prepared by mixing the MTA powder with distilled water in a 1:1 powder/liquid ratio. In the FS group, a sterile cotton pellet dampened with 15.5% ferric sulfate (Astringedent – Ultradent Products Inc, South Jordan, UT, USA) and blotted dry on gauze was placed on the amputated pulp for 15 seconds, followed by the placement of a zinc oxide and eugenol base (Biodinâmica Quím. e Farm. Ltda, Ibiporã, PR, Brazil).

In both groups, a layer of reinforced zinc oxide and eugenol (IRM; Dentsply, Petrópolis, PR, Brazil) was placed prior to the restoration with resin modified glass ionomer cement (RMGIC - Vitremer; 3M ESPE; Sumaré, SP, Brazil). Immediate postoperative periapical radiographies were taken in order to serve as the initial parameter for further postoperative evaluations (5,13).

Clinical and Radiographic Analysis

Periodic follow-up examinations were carried out 3, 6, 12 and 18 months after the end of the treatment. Each checkup involved a clinical and radiographic examination of the pulpotomized teeth, which was performed by two blinded and previously calibrated investigators (kappa values of 0.83 and 0.97 for inter- and intra-examiner reproducibility, respectively).

Clinical success was confirmed in teeth presenting with no spontaneous pain, mobility, swelling or fistula. Radiographic success was considered if internal root resorption, inter-radicular radiolucency and periapical lesion were absent. Hard tissue barrier formation and stenosis were also considered as radiographic successes; tooth discoloration was not considered as a failure.

Histological Analysis

Teeth at the regular exfoliation period were extracted, respecting Nolla stage 8 of the permanent successor, and further processed for histological analysis (13). Following extraction, teeth were immediately immersed in 4% formaldehyde in 0.1M phosphate buffer, pH 7.4, at 4°C for 24 h and in 10% EDTA for 45-60 days. Serial 5-μm-thick longitudinal sections were then prepared, stained with hematoxylin and eosin and viewed under a light microscope. Histologically, the presence of pulp inflammation or even necrosis, intrapulpal calcification, hard tissue barrier, odontoblast layer, and internal resorption were observed and analyzed descriptively.

Statistical Analysis

Clinical and radiographic data were analyzed with generalized estimating equation (GEE). Intra- and inter-examiner reproducibility was determined by Kappa test. The p value ≤0.01 was considered significant.

Results

Thirty-one primary molars from 19 children (13 males and 6 females, ranging from 5 to 9 years, mean age of 7 years and 8 months) were randomly assigned into the two treatment groups: MTA (15 teeth) and FS (16 teeth) by a computer program (Microsoft Excel, Inc, Redmond, WA, USA). At the 18-month follow-up, four teeth (two from each group) were lost due to patient dropout, six teeth from each group had already exfoliated and other six in total were extracted for histological analysis (Fig. 1).

In both groups, 100% of the available teeth were clinically successful during all the follow-up appointments (Fig. 1). No tooth was painful or showed signs of mobility, fistula, swelling or inflammation of the surrounding gingival tissue. The only clinical alteration found in all of the teeth treated with MTA and in two teeth treated with FS was tooth discoloration (p≤0.01), but it was not considered as failure (Fig. 2).

The radiographic success rate for both groups was

100% at 3, 6 and 12 months. At the end of the 18-month follow-up period, one tooth from FS group presented a radiographic failure (inter-radicular radiolucency), but it was not statistically different from MTA group ($p>0.01$). No other radiographic failure, as internal resorption or periapical lesion, was observed (Fig. 1). The radiographic failure was monitored and maintained until the natural exfoliation period since no clinical alteration had been noted.

Radiographically, hard tissue barrier formation was observed only in teeth treated with MTA, which was highly significant ($p\leq0.01$) in comparison with those treated with FS. Regarding radicular stenosis, MTA group had 10 teeth out with root canal obliteration after 12 months and in all of the remaining teeth after 18 months. Only one tooth presented this alteration in the FS group after 3-month follow-up. Considering the radicular stenosis parameter, a statistically significant difference was also found between the groups ($p\leq0.01$). Figure 3 shows the primary mandibular left first and second molars from one patient treated with MTA and FS, respectively, as capping materials in the pulpotomy procedure.

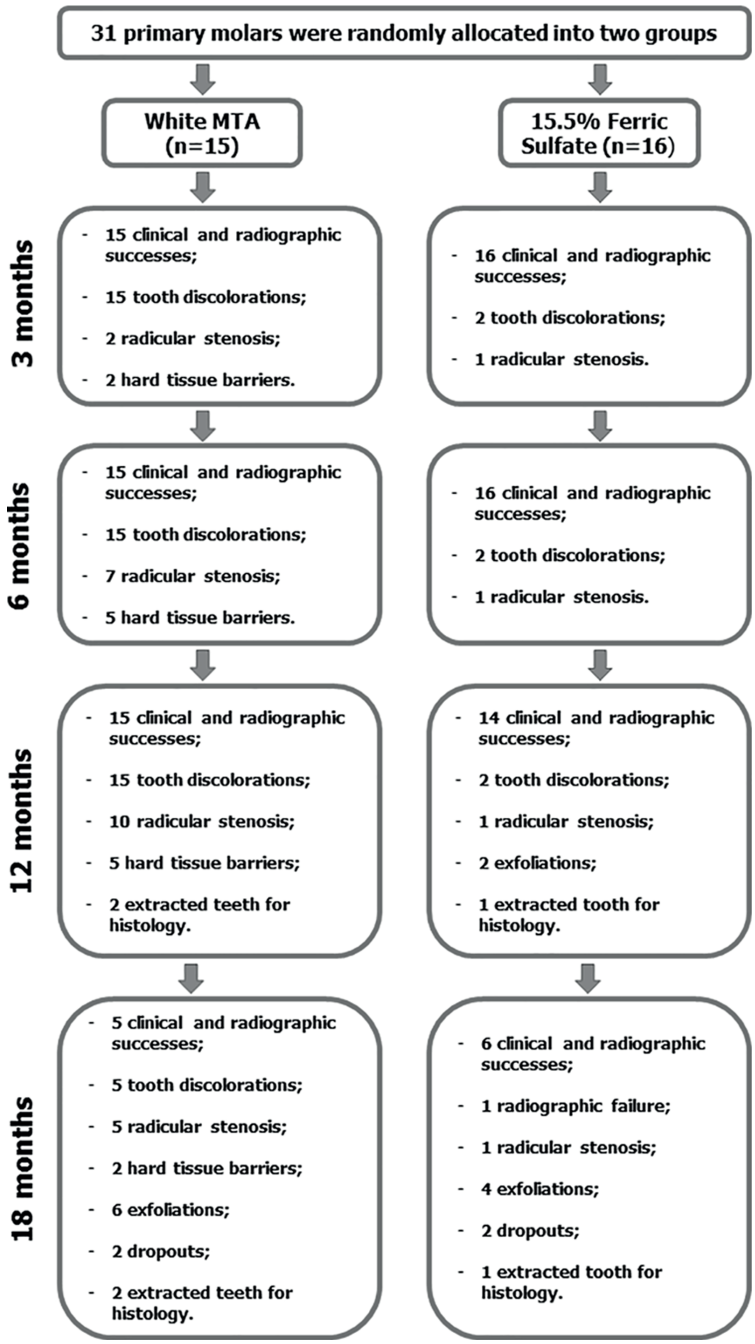

Figure 1. Flow of patients and pulpotomized teeth up to 18 months.

At the end of this study, four teeth from MTA group and two from FS group were evaluated histologically. Tooth extraction occurred around 15 months after the treatment. In general, histological analysis revealed characteristics of pulp vitality and absence of inflammatory infiltrate into the connective tissue of the treated teeth. Dentin-like mineralized tissue deposition was commonly found along the root canals, suggesting root stenosis. MTA group showed hard tissue barrier surrounded by odontoblasts over the pulp stumps, while FS group did not exhibit this condition (Figs. 4 and 5).

Discussion

Pulp injuries due to caries and trauma may threat pulp vitality, so appropriate treatment, such as indirect pulp capping (partial removal of carious dentin), pulpotomy and direct pulp capping must be considered (1). Direct pulp capping is only recommended for primary teeth when a healthy pulp has been accidentally exposed during operative procedures or trauma, and the exposure site is pinpoint in diameter and free of oral contaminants. Pulpotomy is still the most common treatment in cases of cariously exposed pulps in symptom-free primary molars. However, there are increasing evidences that minimally invasive approaches may arrest caries progression allowing the affected primary tooth to remain in place until exfoliation with no pain or infection (14). Therefore, the partial removal of caries stands out as a more conservative procedure than pulpotomy for the treatment of deep carious lesions in teeth with no signs or symptoms of pulp

degeneration. In this procedure, the irreversibly denatured and infected dentine (outer layer) is removed while the affected and capable of being remineralized dentine (inner layer) is preserved (15). Nevertheless, no study has systematically reviewed the existing literature comparing indirect pulp capping, and pulpotomy for primary teeth in children with deep carious lesions and long-term clinical trials are still required to assess the most effective intervention (16).

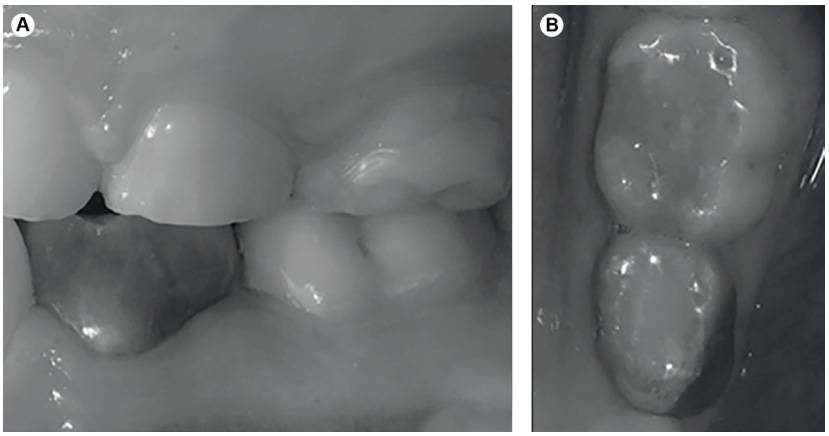

Figure 2. Primary mandibular left first molar treated with MTA presenting tooth discoloration and second molar treated with 15.5% Ferric Sulfate presenting normal tooth color after 18 months. Lateral (A) and occlusal (B) view.

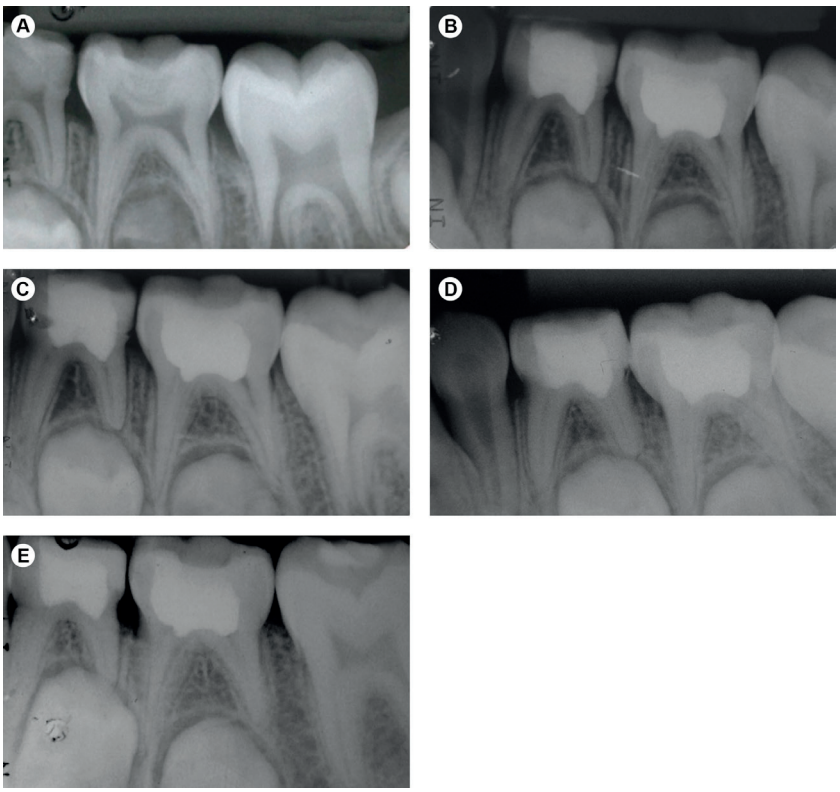

Figure 3. Pulpotomies of primary mandibular left first molar with MTA and second molar with 15.5% Ferric Sulfate (radiographic success). A: Preoperative periapical radiograph of the teeth showing extensive caries lesions, more than two thirds of root lengths, and no signs of periapical lesions; B: 3-month follow-up periapical radiograph showing the pulpotomized teeth; C: 6-month follow-up periapical radiograph suggesting the absence of periapical lesion in the pulpotomized molars; D: 12-month follow-up periapical radiograph suggesting the initial obliteration of the roots and absence of periapical lesion in the pulpotomized molars; E: 18-month follow-up periapical radiograph suggesting the obliteration of the roots and their progressive resorption caused by the eruption of the permanent successors.

To the best of our knowledge, this is the first study evaluating the histological condition of pulp tissue concomitantly with clinical and radiographic features of pulpotomized human primary molars treated with FS and further compared with those treated with MTA. MTA is widely employed for pulpotomies and presents high clinical, radiographic and histological success rates (2,5,6,17). However, its greatest limitation in pediatric dentistry is related to the cost of the material, taking into account the time of survival of the primary tooth in the oral cavity (7). In addition, the routine use of MTA is also limited in developing countries because of economic and commercial reasons (7). FS is an inexpensive alternative to MTA and, although several studies have demonstrated its high clinical and radiographic success rates (4,8-10), few histological studies were performed with FS and these involved the use of animals (12).

Clinically, both MTA and FS were 100% successful over the entire follow-up period. In the literature, the clinical success rates of MTA range between 94% and 100% (2,5,8,10) while those of FS range from 78% to 100%, with the lower rates for the longer periods of evaluation (4,8,10,11).

Radiographically, MTA presented 100% of success during all of the follow-up periods, corroborating the results of other studies, which found radiographic success rates from 94% to 100% in periods of 12 to 74 months (2,8,10,17). For FS group,

radiographic success rate of 100% was also found after 3, 6 and 12 months. At the 18-month follow-up period, the rate decreased to 85.7%. Other authors found lower rates, ranging from 63.3% to 78.6% after 12 months of follow-up (9,10).

Although internal resorption is the most common radiographic failure found in studies with FS (8,9,11,18), it was not observed in none of the teeth assessed in the present work, corroborating the data of Sonmez et al. (4). The occurrence of internal resorption may be attributed to the use of zinc oxide and eugenol (ZOE) as a sub-base, since the eugenol is irritant and may cause a subclinical chronic inflammation (5,10,11). Moreover, other factors are related to this alteration, such as inadequate bleeding control and clot formation, as well as improper restorations with failed sealing, allowing bacterial infiltration and pulp

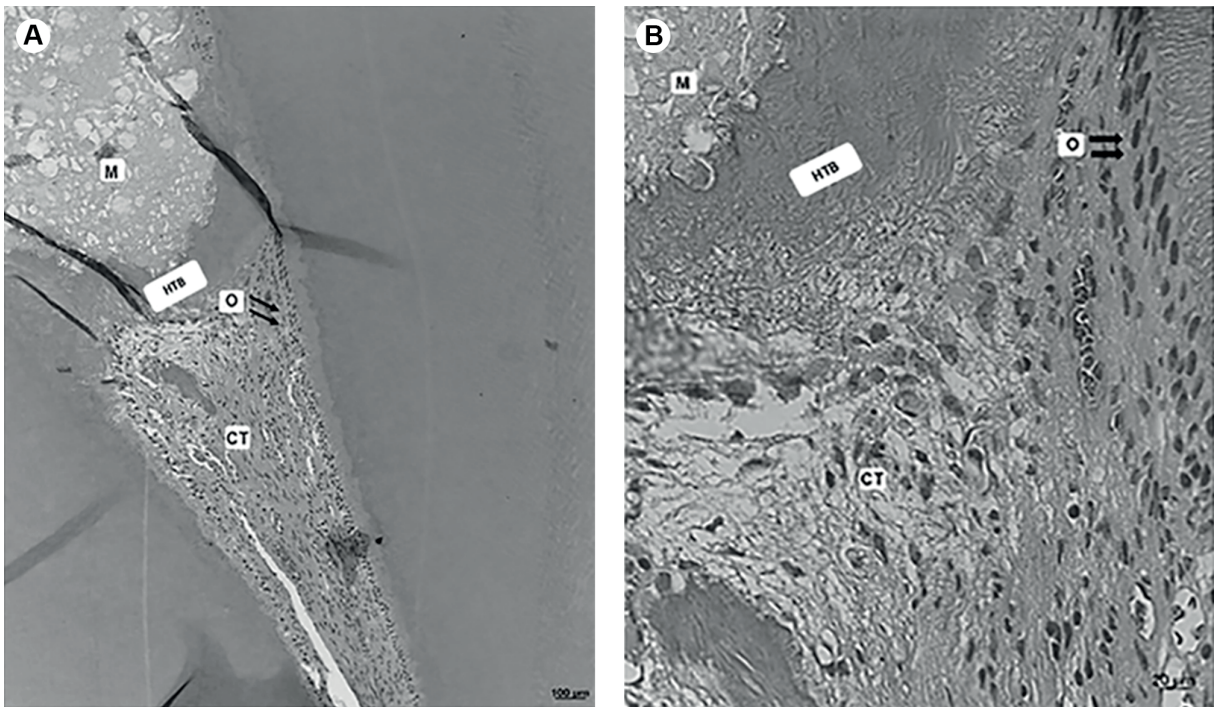

Figure 4. Microscopic aspect of the dentin-pulp complex after pulpotomy with MTA: Sound loose connective tissue (CT), presence of odontoblasts (O) and hard tissue barrier (HTB) between the material (M) and the pulp tissue. Hematoxylin and eosin staining; scale bar of 100 (A) and 20 µm (B).

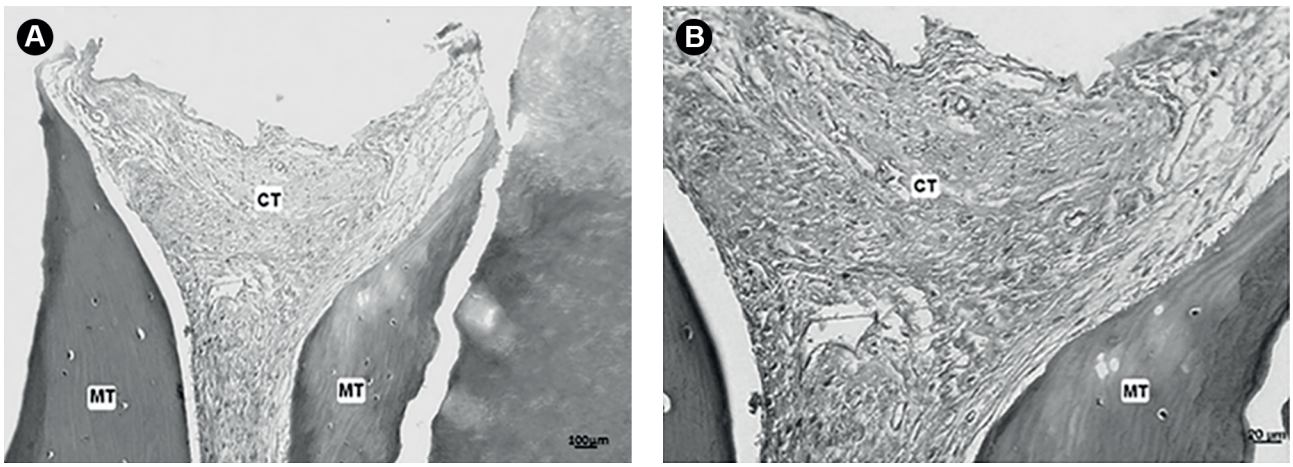

Figure 5. Microscopic aspect of the dentin-pulp complex after pulpotomy with 15.5% Ferric Sulfate: Sound loose connective tissue (CT) with no inflammatory infiltrate, and dentin-like mineralized tissue (MT) deposition in the entire extension of the root canal. Hematoxylin and eosin staining; scale bar of 100 µm (A) and 20 µm (B).

inflammation (4,5). In most of the studies, however, after the use of FS, the pulp stumps were rinsed thoroughly (4,8,10,11). Since we did not perform this procedure, the metal-protein complex formed by FS at the surface of the pulp stumps was not removed and might have acted as a barrier to the irritant components of the sub-base (18) thus impairing the diffusion of eugenol through the pulp tissue. Besides, the formation of this complex prevents the formation of blood clot, thus minimizing the possibility of inflammation and internal resorption (4).

Beyond the capping material properties, the biological sealing after pulpotomy is fundamental for the pulp treatment success (5,19). Final restorations were performed with resin modified glass ionomer cement (RMGIC) instead of composite resin, because our study involved Class I and II cavities of primary molars. According to the recommendations given by American Association of Pediatric Dentistry (AAPD) guidelines, there is strong evidence that composite resins are successful for Class I restoration; for Class II lesions in primary teeth, there is only one randomized controlled trial showing success of composite resin restorations for two years. On the other hand, there is strong evidence that RMGIC for Class I restorations are efficacious, and expert opinion supports Class II restorations in primary teeth (20). A systematic review also supports the use of RMGIC in small to moderate sized Class II cavities (21). Additionally, because of fluoride release, RMGIC may be considered for Class I and II restorations of primary molars in a high caries risk population (22). In this study, the effectiveness of this restorative material, also applied in previous researches (5,13) can be proved by the absence of clinical and almost no radiographic failures.

The only radiographic failure found in one tooth treated with FS was the inter-radicular radiolucency at the 18-month follow-up period. Havale et al. (9) and Fuks et al. (11) also reported this type of failure in similar rates to this study.

The concept of hard tissue barrier is controversial because its presence can be viewed as either a healing response or a pulp reaction to irritation (19). In our study, dentin hard tissue barrier formation was categorized as a radiographic success (2,5,6) and detected in teeth treated with MTA. The presence of hard tissue barrier was further confirmed in the histological analysis, and is attributed to the sealing ability, biocompatibility and alkalinity of the material (8). Hard tissue barrier can present partial or complete formation of mineralized tissue between the capping material and the remaining pulp tissue (19). This structure was neither observed radiographically nor histologically in FS-treated teeth, thus confirming the lack of FS regenerative ability. These data are in agreement with other studies (2,4,5,17).

Radicular stenosis was detected radiographically in 66.66% and 6.25% of the teeth treated with MTA and FS, respectively. Sonmez et al. (4), Erdem et al. (8), Yildiz and Tosun (10) found this alteration respectively in 26.6%, 5% and 21% of the teeth treated with MTA, and in 20%, 5% and 21% of those treated with FS. The active deposition of tertiary reparative dentine and consequently the narrowing of the canals are the result of extensive activity of odontoblast-like cells, thus indicating the presence of pulp vitality (3).

Radiographic evaluation of primary tooth pulpotomies should occur at least annually because the success rate of pulpotomies diminishes over time (23,24). It should be pointed out that, although good radiological practices (use of lead apron, thyroid collars, and high-speed film) were adopted, this study comprised 5 postoperative exposures: one radiograph was taken immediately following the procedure and served as a comparative baseline for the further exposures (3, 6, 12 and 18 months postoperatively). This radiographic control protocol was based on previous studies (5,25). However, excessive exposure is an important issue that must be addressed in future studies since every effort must be made to minimize the patient's exposure.

The histological sections of MTA group showed no inflammation in the entire extension of the root canal and the presence of stenosis and dentin barrier formation. Agamy et al. (17) evaluated the pulp response to white and gray MTA and also found hard tissue barrier in both groups; however, more inflammatory cells and necrotic areas were observed for white MTA, which is in disagreement with our results.

The histological response of pulp tissue to FS was similar to that of teeth treated with MTA, except for the absence of hard tissue barrier. This is not in agreement with the study of Shayegan et al. (12), who observed acute inflammation in most of their samples of primary pig teeth, in addition to necrotic superficial layer and internal resorption in other samples after the treatment with FS. It is worth mentioning that the pattern of necrotic and subjacent inflammatory layers is comparable to that found in teeth treated with formocresol, but without the undesirable features of carcinogenicity and risk of causing enamel defects and abnormal root development of the permanent successor (5,11). Although some authors have noted dentin deposition in the lateral walls of the medium third of the root canal, as seen in present study, some teeth showed incomplete hard tissue barrier formation (12). Our findings show that the use of FS on primary teeth pulpotomy can provide a favorable pulp tissue response.

The present study has the non-compliance and patient dropouts as limitations. Besides, the lack of control over the

tooth extraction period results in the difficulty to obtaining the histological slides (13). Even though our results are encouraging, the number of teeth assessed histologically was obviously small and cannot be extrapolated to general population. Further long-term follow-up studies are needed to evaluate the histological reaction of the dental pulp mainly to FS.

Based on this study, both MTA and 15.5% FS are effective for pulpotomies of primary teeth. Although MTA is considered the first choice material, FS may be a suitable alternative when treatment cost is an issue.

Resumo

O objetivo deste estudo foi avaliar os resultados clínicos, radiográficos e histológicos do complexo dentino-pulpar de molares decíduos após pulpotomia com agregado trióxido mineral (MTA) e sulfato férrico (SF) 15,5%. Trinta e um molares decíduos foram alocados aleatoriamente nos grupos MTA ou SF. As avaliações clínicas e radiográficas foram realizadas aos 3, 6, 12 e 18 meses de acompanhamento. Os dentes no período regular de esfoliação foram extraídos e processados para análise histológica. Os dados clínicos e radiográficos foram analisados estatisticamente (p≤0,01). O resultados histológicos foram analisados descritivamente. Todos os dentes tratados apresentaram sucesso clínico ao longo dos períodos experimentais. Ambos os grupos exibiram 100% de sucesso radiográfico aos 3, 6 e 12 meses. Aos 18 meses de acompanhamento, um dente do grupo SF apresentou radiolucidez interradicular (p>0,01). Histologicamente, os dentes tratados apresentaram vitalidade pulpar e ausência de infiltrado inflamatório no tecido conjuntivo. Somente o grupo MTA mostrou barreira de tecido duro rodeada por odontoblastos sobre os cotos pulpares. MTA e SF 15,5% são eficazes para pulpotomias de dentes decíduos. Embora o MTA seja considerado o material de primeira escolha, o SF pode ser uma alternativa adequada quando o custo do tratamento é um problema.

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