

case report

Outcomes of an alternative cervical sealing material in traumatized immature teeth subjected to pulp revascularization: a case series

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Abstract

Introduction and objective: Pulp revascularization is a viable and effective treatment for immature necrotic teeth. The purpose of this case series was to report the outcomes of pulp revascularization using an alternative cervical sealing material (composed by calcium hydroxide, 2% chlorhexidine gel, and zinc oxide), which showed an acceptable clinical and radiographic performance in further indications, such as root canal filling and root resorptions. Case report: We selected seven traumatized immature permanent maxillary central incisors with crown fracture, extrusive luxation, and pulp necrosis. For pulp revascularization, we followed four main steps: passive decontamination, three-week interappointment dressing, scaffold, and cervical sealing. The cervical sealing was carried out with the association of calcium hydroxide, 2% chlorhexidine gel, and zinc oxide in a proportion of 2:1:2. The follow-up period was 24 months. We classified pulp revascularization outcomes as complete healing, incomplete healing, or failure. All cases showed complete healing.

Furthermore, in all cases, the proposed cervical sealing material remained stable throughout the whole follow-up period. **Conclusion:** The material evaluated did not induce crown discoloration in the study sample. Therefore, the material composed of a mixture of calcium hydroxide, 2% chlorhexidine gel, and zinc oxide can be an alternative for cervical sealing material in pulp revascularization.

Introduction

Pulp revascularization may be considered the first treatment option for immature teeth with pulp necrosis, as it shows favorable clinical outcomes such as radiographic evidence of continued root development [4]. The biological basis of this therapy is that it is effective for the decontamination of the root canal system and promotes stem cell differentiation of apical papilla. It is also effective for obtaining an appropriate scaffold for the regulation of cell differentiation and, finally, placing a cervical barrier over the scaffold using a biocompatible sealing material [4, 13].

The cervical barrier/sealing is an important step in pulp revascularization, since it promotes a physical barrier that prevents the root canal recontamination. The sealing material should prevent bacterial or toxin leakage and allow cell proliferation and differentiation [21]. Therefore, this process must be accomplished with a biocompatible material, once it is applied over the scaffold.

The most widespread protocol regarding the sealing of pulp revascularization is the use of mineral trioxide aggregate (MTA) in the cervical third of the root canal [13, 14]. Few studies in the literature use other alternatives for this purpose, e.g., EndoSequence Root Repair Material (Brasseler, Savannah, GA, United States), Biodentine (Septodont, Saint-Maur-des-Fosses, Paris, France), IRM, and glass ionomer [14]. MTA induces the production of an apatite layer when in contact with phosphate-containing physiological fluids in a process called biomineralization [9]. This material can also support human mesenchymal stem cell adhesion, proliferation, and migration [21]. However, MTA presents some limitations, such as the induction of crown discoloration, which is critical for coronary and cervical root areas [4, 5].

Moreover, a material composed of a mixture of calcium hydroxide, 2% chlorhexidine gel, and zinc oxide has been used as an intracanal dressing, without periodic changes in cases of traumatized immature teeth, for apexification in a single-visit approach [2]. It has shown clinical and radiographic success with apical barrier formation, control of root resorptions and dimensional long-date stability on previous clinical evaluations [2, 10, 16, 18].

Therefore, the purpose of this case series was to report the outcomes of pulp revascularization using an alternative cervical sealing material (composed by calcium hydroxide, 2% chlorhexidine gel, and zinc oxide).

Case report

Sample selection

This study was approved by the local ethics research committee (46653015.0.0000.5418). All patients were informed about the possible risks and benefits of the treatment and signed the informed consent form. For the research, we selected seven teeth from five patients (7–15 years of age; mean age of participants \pm standard deviation = 10 \pm 2.5). Patients received treatment between 2015 and 2017 at the University Dental Trauma Service, by an endodontist with more than five years of experience. We also obtained detailed medical and dental trauma history, demographic data, and clinical and radiographic characteristics. Specific cases that fulfilled the following restricted criteria were included in the outcome analysis:

- Immature permanent maxillary central incisors with crown fracture and extrusive luxation;
- Pulp necrosis resulting from dental trauma, with or without clinical signs and symptoms or periapical radiolucency;
- A tooth treated with the pulp revascularization protocol described in this study by an endodontist.

Pulp revascularization protocol

We used the pulp revascularization protocol, according to Nagata *et al.* [15], with modifications. Clinical examination included the presence of spontaneous pain, tenderness to percussion, pain on palpation, and sinus tract, mobility, swelling or crown discoloration. The diagnosis of pulp necrosis was obtained with pulp sensibility test (EndoFrost[™] Roeko, Langenau, Germany) and by electric pulp test (Pulp Tester Digital Odous de Deus, Belo Horizonte, MG, Brazil). We also applied infiltrative anesthesia with 2% lidocaine and 1:100,000 epinephrine (Alphacaine; DFL, Rio de Janeiro, RJ, Brazil).

For the tooth field isolation, we used a rubber dam without clamps, and to disinfect the operative field, 2% chlorhexidine gel on a cotton swab. Then, the access cavity was prepared using a diamond bur (KG Sorensen[™], Barueri, SP, Brazil) with copious irrigation. The teeth full length was initially determined with Apixia digital imaging software (Apixia, Industry, CA, United States), but the length was established 4 mm short of the total root length due to the injection of chemical solutions.

The passive decontamination protocol was used for root disinfection. Slow and careful irrigation was obtained with 10 mL of 6% sodium hypochlorite (Drogal, Piracicaba, SP, Brazil), followed by inactivation with 10 mL of 5% sodium thiosulfate (Drogal, Piracicaba, SP, Brazil) and saline solution. Then, we applied 10 mL of 2% chlorhexidine (Drogal, Piracicaba, SP, Brazil), that was neutralized by 5% Tween 80, 0.07% soy lecithin (Drogal, Piracicaba, SP, Brazil) and a saline solution. The root canal was dried with the aid of a capillary tip (Ultradent Products, South Jordan, UT, United States) 3 mm short of the total root length.

After these procedures, a three-week interappointment dressing was applied. For this, root canals were filled with a paste composed of calcium hydroxide (Biodinâmica, Ibiporã, PR, Brazil) and 2% chlorhexidine gel (Endogel, Itapetininga, SP, Brazil) as a vehicle, at the proportion of 1:1, inserted with a lentulo spiral (Dentsply Maillefer[™], Tulsa,

OK, United States). Then, the teeth were sealed with an inner layer of Coltosol (Coltène/Whaledent[™], Mahwah, NJ, United States), approximately 2-mm thick, and an external layer of composite resin (Filtek 3M ESPE[™], Sumaré, SP, Brazil). In the second appointment, the intracanal dressing content was removed with 10 mL of saline solution.

All cases received final irrigation with 3 mL of 17% EDTA (Fórmula e Ação, São Paulo, SP, Brazil) accomplished for 3 minutes, followed by irrigation with saline solution. The root canal space was then dried with the aid of a capillary tip (Ultradent Products, South Jordan, UT, United States) and sterile paper points. To induce bleeding into the root canals, we placed a #35 manual k-file (Dentsply Maillefer, Ballaigues, Switzerland) at 1-2 mm beyond the apical foramen. A collagen matrix (CollaCote; Zimmer Dental, Carlsbad, CA, United States) was inserted above the blood clot, and then a cervical sealing was also applied.

Cervical sealing protocol

Cervical sealing was performed with the association of calcium hydroxide, 2% chlorhexidine gel, and zinc oxide (S.S. White Artigos Dentários LTDA, Rio de Janeiro, RJ, Brazil), in the proportion of 2:1:2, respectively (Figure 1). After handling, the consistency of the paste was similar to the one of Coltosol temporary filling material (Coltène/Whaledent[™], Mahwah, NJ, United States). The material was applied above the cementoenamel junction with the aid of an endodontic condensor. The access cavity was subsequently double sealed with Coltosol and composite resin, as previously described.

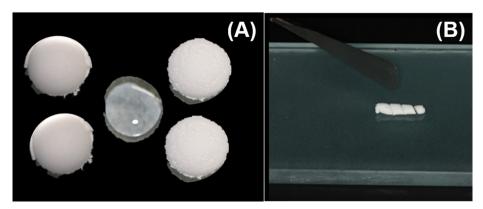


Figure 1 – Alternative cervical sealing material proposed. (A) Components of the material: calcium hydroxide, 2% chlorhexidine gel and zinc oxide, in the proportion of 2:1:2, before handling. (B) Consistency of the material after adequate handling

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Follow-up and outcomes analysis criteria

All patients were subjected to follow-up every three months for 24 months. For clinical and radiographic outcome analysis, initial exams were compared to the data collected after the entire follow-up period. Postoperative clinical aspects - spontaneous pain, tenderness to percussion, pain on palpation, and presence of sinus tract, mobility, swelling or crown discoloration - were also considered. The endodontist visually assessed crown discoloration, and compared the treated tooth with adjacent teeth under ambient lighting. In addition, the pulp sensibility test and electric pulp test were also performed, as previously described. The preoperative and postoperative radiographic examination included: the presence of periapical radiolucency and of root resorption, Cvek's stage of root development [1], and stability of cervical sealing. Two experienced endodontic specialists performed the radiographic analysis.

For the analysis of pulp revascularization outcome of the cases, we used the study by Jeeruphan *et al.* [11] with modifications, presenting one of the three classifications, as follows:

- Complete healing, when presenting:

- No clinical signs and symptoms;
- Complete resolution of the initial periradicular radiolucency, if applicable;
- Stabilization of initial external inflammatory root resorption, if applicable.
- Incomplete healing, when presenting:
 - No clinical signs and symptoms;
 - Incomplete resolution of the initial periradicular radiolucency, if applicable;
 - Stabilization of initial external inflammatory root resorption, if applicable.
- Failure (disease), if presenting:
 - Clinical signs and symptoms;
 - Progression of the initial periradicular radiolucency;
 - Progression of initial external inflammatory root resorption.

Two experienced evaluators (endodontists) analyzed the outcomes until reaching a consensus. The results were then tabulated and analyzed, and the prevalence (%) of the data was obtained.

Results

Table I summarizes the patients' demographic and initial clinical and radiographic information (teeth included). Regarding follow-up and outcomes analysis, the results are expressed in Tables II and III, respectively. All cases showed complete healing, and no tooth showed incomplete healing or treatment failure. A total of four teeth (57%) responded to the electric pulp test in the 24 months of follow-up.

Concerning cervical sealing, the proposed material remained stable radiographically throughout the whole period of 24 months in all cases (Figure 2). Moreover, the teeth presented no crown discoloration after pulp revascularization, regardless of the cervical sealing height, which was observed at the cervical root region in most cases (72%), followed by the middle root third (28%).



Figure 2 – Representative periapical radiograph images of complete healing and cervical sealing adequacy after a two-year-follow-up. This case involved an 8-year-old male who traumatized the right and the left central maxillary incisors after a fall (causing enamel-dentin-fracture associated with extrusive luxation). **(A)** Preoperative radiograph showing periapical radiolucency. The patient had not reported clinical symptoms. **(B)** Postoperative radiograph after pulp revascularization. **(C)** Threemonth-follow-up. **(D)** Six-month-follow-up. **(E)** Twelvemonth-follow-up. **(F)** Twenty-four-month-follow-up showing an absence of apical closure and an absence of continued root development

 Table I - A summary of patients' demographic data and the initial clinical and radiographic characteristics of the teeth selected in this study*

Variable	Number of teeth (frequency)
Gender	
Male	5 (72%)
Female	2 (28%)
Age (years)	10 ± 2.5
Type of trauma	
Enamel fracture + extrusive luxation	2 (29%)
Enamel-dentin fracture + extrusive luxation	4 (57%)
Enamel-dentin-pulp fracture + extrusive luxation	1 (14%)
Etiology	
Fall	4 (57%)
Sports	2 (29%)
Bicycle	1 (14%)
Preoperative clinical symptoms	4 (57%)
Preoperative periradicular radiolucency	5 (71%)
Preoperative root resorption	2 (29%)
Initial Cvek's stage of root development	$2.9~\pm~0.7$

*Age and initial Cvek's stage of root development are represented as mean and standard deviation

Table II - Clinical and radiographic findings after a two-year-follow-	Table	II - C	Clinical	and	radiogra	ohic	findinas	after a	two-	vear-follow-u
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Variable	Number of teeth (frequency)
Apical closure	
Yes	3 (43%)
No	4 (57%)
Continued development in root width and length	1
Yes	0 (0%)
No	7 (100%)
Blood clot height	
Apical third	0 (0%)
Middle third	5 (71%)
Cervical third	2 (29%)
Final Cvek's stage of root development*	4 ± 1
Response to pulp sensibility test during follow-u	p
Yes	0 (0%)
No	7 (100%)
Response to electric pulp test during follow-up	
Yes	4 (57%)
No	3 (43%)

*Final Cvek's stage of root development is represented as mean and standard deviation

Treatment outcomes		Number (%)	
	Complete healing	7 (100) 0 (0)	
Pulp revascularization outcomes	Incomplete healing		
	Failure (disease)	0 (0)	
	Total	7 (100)	

Table III - Number and frequency of cases obtained according to established treatment success

Discussion

Few studies in the literature address the most indicated treatment protocols and materials, as well as the long-term treatment efficacy [20]. In this study, the objective was to report the outcomes of an alternative cervical sealing material in pulp revascularization that does not induce crown discoloration and which could also turn this treatment more accessible.

The proposed material is composed of calcium hydroxide, 2% chlorhexidine gel, and zinc oxide, three components largely evaluated in the endodontic literature. Calcium hydroxide is one of the most used medications in dentistry, mainly as an intracanal dressing for teeth with incomplete root formation [17]. It induces the formation of mineralized tissue and presents antimicrobial activity due to its alkaline pH [6, 17]. The second component, chlorhexidine, has a high antimicrobial activity, substantivity (residual antimicrobial activity acting from one to 12 weeks) and biocompatibility [8]. Therefore, the main goal of associating calcium hydroxide with 2% chlorhexidine gel is to enhance the antimicrobial effectiveness of the material, mainly against microorganisms resistant to an alkaline environment [3, 7, 17].

Concerning zinc oxide, it is a water- and serum-resistant component that allows for adequate consistency and radiopacity of the material [3]. We emphasize that the use of the proposal material was based on previous clinical studies that had acceptable outcomes when applied in further indications such as root canal filling and root resorptions as an obturator material [10, 16]. In a rat subcutaneous tissue reaction study, this material showed favorable results for biocompatibility [19].

Furthermore, the association between these three components has satisfactory antimicrobial activity, even in deep regions of dentinal tubules [3, 7]. Considering the availability of the material components, it presents a relatively low cost, being relevant in third world countries.

In this case series, we detected no treatment failure, and stable sealing was observed through the

whole period of follow-up. A relevant feature is the increase in root length and thickness of dentinal walls, but in this study these desirable outcomes could not be reached. The sample presented a severe trauma injury with crown fracture and extrusive luxation, which may have interfered with the viability of mesenchymal stem cells that are important for the healing process. Other features that could yield minor and/or late root development is the preoperative presence of periradicular radiolucency, clinical symptoms and root resorption, observed in 71, 57 and 29% of the patients in this study, respectively. However, we observed regression of all signs and symptoms of infection in the patients after pulp revascularization.

As a result, all cases presented positive outcomes similar to the ones of other cervical sealing materials with clinical and radiographic success. The proposed material composition thus allowed to create a suitable root environment for pulp revascularization and showed dimensional long-term stability. The material seems to be an interesting alternative for cervical sealing in pulp revascularization, as it has relatively low cost, it is easy to use, available in the market and it does not induce crown discoloration. The association of calcium hydroxide, 2% chlorhexidine gel and zinc oxide has already been used as revascularization cervical sealing in a recent case report that demonstrated consistent results with this study [16].

Crown discoloration, a considerable adverse effect in pulp revascularization, was not observed in our study. This is a matter of concern since most materials used in pulp revascularization have the potential to stain tooth tissues, as recently confirmed by a systematic review [5]. MTA, the most common material used as cervical sealing, has been associated with crown discoloration in several pulp revascularization studies [4, 5, 12]. OrthoMTA and RetroMTA have also been associated with unaesthetic outcomes [5]. The proposed material here did not induce crown discoloration in the sample. Despite that, the excess coronal bleeding must always be carefully removed, so that it remains above the cementoenamel junction. The contact of the cervical sealing material with bleeding may also be associated with crown discoloration [5].

We suggest the use of a periodontal probe to measure the tooth crown extension and to ensure the adequate height of the blood clot within the root cavity. In addition, the placement of a collagen membrane below cervical sealing is of utmost importance for better results [13]. The membrane absorbs moisture from the blood clot, avoiding contact between the cervical sealing material and blood, and allows for the adequate material deposition, minimizing the risk of displacement or resorption [4].

The return of pulp sensibility after pulp revascularization occurs in approximately 50% of the cases [4]. The presence of functional nociceptors following the pulp revascularization suggests that apically free nerve endings are guided into the canal by specific chemical signals [4]. In this study, four teeth (57%) responded to the electric pulp test during the follow-up visits, including teeth that had preoperative radiolucency or signs and symptoms. Furthermore, from this sample of four teeth, the majority (75%) presented the blood clot height in the cervical region, probably due to a greater amount of blood that filled the pulp space after bleeding stimulation.

Conclusion

The material evaluated did not induce crown discoloration in the study sample. Therefore, the material composed by a mixture of calcium hydroxide, 2% chlorhexidine gel, and zinc oxide can be an alternative cervical sealing material in pulp revascularization.

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References

Cvek M. Prognosis of luxated non-vital maxillary incisors treated with calcium hydroxide and filled with gutta-percha. A retrospective clinical study. Endod Dent Traumatol. 1992 Apr;8(2):45-55.

de Jesus Soares A, Yuri Nagata J, Casarin RC, Almeida JFA, Gomes BP, Zaia AA, et al. Apexification with a new intra-canal medicament: a multidisciplinary case report. Iran Endod J. 2012;7(3):165-70.

de Souza-Filho FJ, Soares A de J, Vianna ME, Zaia AA, Ferraz CC, Gomes BP. Antimicrobial effect and pH of chlorhexidine gel and calcium hydroxide alone and associated with other materials. Braz Dent J. 2008;19(1):28-33.

Diogenes A, Ruparel NB. Regenerative endodontic procedures: clinical outcomes. Dent Clin North Am. 2017 Jan;61(1):111-25.

Fagogeni I, Metlerska J, Lipski M, Falgowski T, Maciej G, Nowicka A. Materials used in regenerative endodontic procedures and their impact on tooth discoloration. J Oral Sci. 2019 Aug;61(3):379-85.

Gomes BP, Ferraz CC, Vianna ME, Rosalen PL, Zaia AA, Teixeira FB, et al. In vitro antimicrobial activity of calcium hydroxide pastes and their vehicles against selected microorganisms. Braz Dent J. 2002;13(3):155-61.

Gomes BP, Montagner F, Berber VB, Zaia AA, Ferraz CC, de Almeida JF, et al. Antimicrobial action of intracanal medicaments on the external root surface. J Dent. 2009 Jan;37(1):76-81.

Gomes BP, Vianna ME, Zaia AA, Almeida JF, Souza-Filho FJ, Ferraz CC. Chlorhexidine in endodontics. Braz Dent J. 2013;24(2):89-102.

Guimarães BM, Prati C, Duarte MAH, Bramante CM, Gandolfi MG. Physicochemical properties of calcium silicate-based formulations MTA Repair HP and MTA Vitalcem. J Appl Oral Sci. 2018;26:e2017115.

Herrera DR, Herrera CM, Lima AR, Nagata JY, Pereira AC, Silva EJ, et al. Repair of apical root resorption associated with periodontitis using a new intracanal medicament protocol. J Oral Sci. 2014 Dec;56(4):311-4. Jeeruphan T, Jantarat J, Yanpiset K, Suwannapan L, Khewsawai P, Hargreaves KM. Mahidol study 1: comparison of radiographic and survival outcomes of immature teeth treated with either regenerative endodontic or apexification methods: a retrospective study. J Endod. 2012 Oct;38(10):1330-6.

Kahler B, Rossi-Fedele G. A review of tooth discoloration after regenerative endodontic therapy. J Endod. 2016 Apr;42(4):563-9.

Kim SG, Malek M, Sigurdsson A, Lin LM, Kahler B. Regenerative endodontics: a comprehensive review. Int Endod J. 2018 Dec;51(12):1367-88.

Lee JY, Kersten DD, Mines P, Beltran TA. Regenerative endodontic procedures among endodontists: a webbased survey. J Endod. 2018 Feb;44(2):250-5.

Nagata JY, Gomes BP, Rocha Lima TF, Murakami LS, de Faria DE, Campos GR, et al. Traumatized immature teeth treated with 2 protocols of pulp revascularization. J Endod. 2014 May;40(5):606-12.

Prado MC, Cerqueiro-Neto ACCL, Secchi P, Pereira AC, Gomes BPFA, Soares AJ. Recurrence of dental trauma and management of pulp revascularized tooth: a case report. J Dent Health Oral Disord Ther. 2018;9(4):304-18.

Signoretti FG, Gomes BP, Montagner F, Barrichello Tosello F, Jacinto RC. Influence of 2% chlorhexidine gel on calcium hydroxide ionic dissociation and its ability of reducing endotoxin. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2011 May;111(5):653-8.

Soares AJ, Lima TFR, Nagata JY, Gomes BPFA, Zaia AA, Souza-Filho FJ. Intracanal dressing paste composed by calcium hydroxide, chlorhexidine and zinc oxide for the treatment of immature and mature traumatized teeth. Braz J Oral Sci. 2014 Jan/Mar;13(1):6-11.

Soares AJ, Prado M, Brazão MA, Gomes BPFA, Zaia AA. The biocompatibility of a new endodontic paste used in dental trauma. Rev Odontol UNESP. 2015 July/Aug;44(4):232-8.

Torabinejad M, Nosrat A, Verma P, Udochukwu O. Regenerative endodontic treatment or mineral trioxide aggregate apical plug in teeth with necrotic pulps and open apices: a systematic review and metaanalysis. J Endod. 2017 Nov;43(11):1806-20.

Wongwatanasanti N, Jantarat J, Sritanaudomchai H, Hargreaves KM. Effect of bioceramic materials on proliferation and odontoblast differentiation of human stem cells from the apical papilla. J Endod. 2018 Aug;44(8):1270-5.