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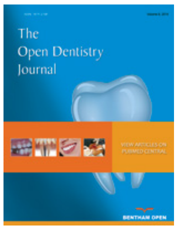

REVIEW ARTICLE

Pulp Revascularization: A Literature Review

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Abstract: Reestablishing blood flow and allowing the continuation of root development are some of the objectives of pulp revascularization. This procedure is currently indicated for teeth with incomplete root formation as an alternative to the traditional treatment of apecification, which consists of inserting calcium hydroxide paste into the root canal for a determined time period in order to induce the formation of a calcified barrier. Although it is considered as the most classically employed therapy, the permanence of the paste for long time periods may lead to the weakening of the root due to hygroscopic properties, as well as proteolytic activities of calcium hydroxide. Therefore, there has been a permanent search for alternatives which allow the full development of immature teeth. Revascularization has emerged as such an alternative, and a range of treatment protocols can be found in the scientific literature. The aim of this paper is to accomplish a literature review concerning this issue.

Keywords: Endodontics, therapeutics, calcium hydroxide, blood vessels.

INTRODUCTION

Conventional endodontic treatment consists in the development and application of techniques designed to accomplish the chemical-mechanical preparation of root canals in order to eliminate an infection, many times difficult to combat due to the complexity of the root canal system. However, this process may become even more complicated in cases of immature teeth with open apexes, whose root walls are fragile due to the thin thickness of the root canal dentin, along with the intense activity and anatomy of an open apex, making it difficult to accomplish the complete obturation of the canal, and with the real risk of solid and plastic material overflow into the periapex. The incomplete root development may be caused by trauma or infections powerful enough to halt mineral deposition by the destruction of blood flow, impeding the root to complete its formation [1].

One way to treat open apex teeth is the apexification technique that is made in pulpless teeth and which promotes apical closure, and can be obtained with the insertion of a MTA (Mineral Trioxide Aggregate) barrier or with periodical exchanges of calcium hydroxide, enhancing further obturation. This process is named apexogenesis and its goal targets at the preservation of vital pulp tissue so that the continued root development with apical closure may occur [2, 3]. Nevertheless, some studies have shown that this protocol can also be used in non-vital teeth. The procedure is named pulp revascularization and is directed by disinfection protocols concerning the root canals, being indicated the use of sodium hypochlorite irrigation (NaOCl) followed by a combination of ciprofloxacin, metronidazole and minocycline antibiotics to be used as intracanal medication [4].

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The aim of this work is to accomplish a literature review concerning pulp revascularization and its efficacy as a Regenerative Endodontic Procedure.

LITERATURE REVIEW

The dental pulp consists mainly of a loose connective tissue with a variety of specialized cells such as odontoblasts mixed with more commonly found other cells like fibroblasts, endothelial cells, nerve cells, immune cells, and more recently discovered stem/progenitor cells, along with an extracellular matrix including fibrillar proteins and ground substance, making the dental pulp a unique organ, being an adult person able to have a total of up to 52 pulp organs [5]. Bacterial, mechanical or physic-chemical factors are powerful enough to harm the pulp, leading to vascular changes and inflammation, the pain being described as execrating and almost intolerable, and making the patients search for dental help.

The first attempts to regenerate the pulp tissue were conducted by Nygaard Ostby [6, 7]. In both studies, the root canals were intentionally overinstrumented to evoke bleeding followed by the obturation with gutta-percha and Kloroperka N-O paste short of the root apices to allow tissue ingrowth into the root canal space. They also used a 4% formaldehyde solution to disinfect the canals in cases where necrosis was present. The histological examinations of these showed mineral tissue deposition along the root canal walls and connective tissue.

With the importance of infection control in mind and considering the conditional role of microorganisms within the root canals, Rule and Winter [8] introduced polyantibiotics consisting of a mixture of neomycin sulfate, polymyxin B sulfate, bacitracin, and also nystatin, associated with absorbable iodoform into the root canals, which caused a thicker and continued root development as well as an apical barrier formation in pulpless teeth. As the researches went on, Nevins and collaborators [9, 10] reported revitalization and hard tissue formation in immature pulpless teeth in monkeys and humans when root canals were mechanically instrumented and collagen–calcium phosphate gels were used as a scaffold.

Although the presence of hard tissue formation began to be discussed in the dental community, it was also observed that the teeth treated with this therapy were more prone to fracture under stress due to the thin dentin walls, and so it became only natural and expectable that scientists would find out a way to stimulate the organism to complete root development, including the apex closure, opening the era of Regenerative Endodontic Procedures (REP), designed to predictably replace damaged, insufficient and missing structures by healthy newly produced tissues, restoring the shape and function of the pulp-dentin complex. The new approaches included: direct pulp capping, revascularization, apexogenesis, apexification. Stem cell therapy, and tissue engineering are the most up to date ones [11]. Most of the therapies mentioned above use the host´s own vascular cells, and two modalities of treatment became more clinically available: apexification and revascularization; the former using calcium hydroxide, which requires a long time for the treatment, and MTA which neither strengthens the root nor promotes further development; the latter by promoting further root development resulting in the reinforcement of the dentinal wall by the deposition of hard mineral tissue, strengthening the roots.

Regenerative Endodontic Procedures so have emerged as a viable, easy doing alternative to allow the complete formation of the roots of immature teeth, mainly in the last decade. REPs work with the prerogative that the root canal space free of contamination associated with a new stimulated blood supply can indeed reestablish vascularization, enhancing root completion the gap between the theory and clinical applications has been narrowed, and researches are now converging to regenerative procedures [12, 13].

Only in 2008 was pulp revascularization procedure globally available in the internet by Trope [14] who described it and applied the technique in a lower right second premolar with open apex, with clinical and radiographic aspects of apical periodontitis, with the presence of a fistule. Irrigation was accomplished with 5.25% sodium hypochlorite and a blood clot was produced at the level of the cementum to give structural support for new tissue growth, followed by a double sealing with MTA in the cervical region, and then restored with composite resin. Clinical and radiographic healing could be observed after 22 days. The author claimed that if revascularization is not reached in a period of three months, conventional treatment is then indicated.

Kvinnsland [15] reported a case of a 9-year-old patient with history of trauma in the upper central incisors. After clinical and radiographic examination, the diagnosis of concussion was obtained and emergency dental assistance took place. A month later the patient reported slight symptoms in the area, being diagnosed with periapical periodontitis. The root canal was then instrumented and irrigated with 0.5% sodium hypochlorite and filled with a paste of calcium hydroxide and IRM, along with the accomplishment of weekly periapical radiographs. Four months later, root formation began to be visualized, and the exchange of intracanal calcium hydroxide was made, and periodical radiographs were accomplished every 3 months, which revealed continuous root formation and apical closure. The authors reported that the kind of pulp response varies, not only according to the type of the traumatic injury, but also concerning the action of progenitor cells implied in the process. Still according to them, tissue repair may be initiated from pulp progenitor cells, periodontal tissues or by the combination of both. If the damaged pulp tissue is renewed by pulp progenitor cells, there seems to be repair by the action of dental pulp stem cells (DPSCs) which can be induced, at least *in vitro*, to differentiate into odontoblastic phenotype, characterized by polarized cells and mineralized nodules [16, 17], while producing newly formed dentin which can be reinnervated by sensorial nerves.

Shimizu [18] defined regeneration as the replacement of damaged tissues by the same type of parenchyma cells previously existent in that tissue. With the aim of scrutinizing histological aspects of a tooth with incomplete root formation and diagnosed with irreversible pulpitis, the authors described a case report of a 10-year-old boy who underwent revascularization therapy and showed normal periapical tissues after the treatment, accomplished with a MTA barrier. However, the tooth was fractured soon after and needed to be extracted. After having been stained following hematoxylin/eosin protocols, connective loose tissue with few collagen fibers were observed filling the root canal space as far as the MTA barrier, while the pulp tissue as a whole did not show inflammatory cells. The majority of the cells in the root canal space, especially in the periapical area, were classified as fusiform cells, fibroblasts or mesenchymal cells, with more blood vessels and cells in the root canal than in the apical zone. The authors concluded that revascularization and regeneration of a permanent tooth with incomplete root formation and irreversible pulpitis, involving the pulp tissue in the apical portion of the root canal may indeed have the potential of pulp tissue regeneration.

In the study of Pramila and Muthu [19], the authors reported the results of treatments in patients with incomplete root formation of permanent teeth, with and without pulp vitality. The vital teeth, after instrumentation, were filled with antibiotic paste and sealed with IRM® zinc eugenol temporary sealer. In the following section, clot formation was stimulated through overinstrumentation followed by MTA® insertion and sealing with IRM®. Twenty four hours later the teeth were sealed with glass ionomer (GIC). In necrotic teeth, after disinfection, clot formation was stimulated. The authors concluded that under certain circumstances, teeth with necrotic pulps and open apexes are able to regenerate pulp tissue and to promote hard tissue production associated with root growth and complete apex formation.

Aggarwal [20], in 2012, compared the apexification accomplished with calcium hydroxide and pulp revascularization in a single 24-year-old female patient, but in different teeth. The patient complained of pain, edema and mobility in the upper central incisors and purulent discharge in the frontal upper region of the face. She gave a history of trauma around 15 years before, and her medical history was noncontributory. In the radiographs, incomplete rhyzogenesis could be observed along with thin radicular walls. Both the central incisors had grade I mobility and the intraoral radiographs also revealed immature apices associated with both maxillary central incisors, but with no signs of fracture despite the fact that the walls of the canals were very thin. Irrigation was made with 5.25% sodium hypochlorite and instrumented. Calcium hydroxide paste was inserted in the root canal of tooth 11, while in tooth [21], a three antibiotic creamy paste and provisory sealing with zinc eugenol sealer took place. Two weeks later, these procedures were repeated, and after two months tooth 11 were obturated with guttapercha, while tooth 21 underwent revascularization procedure, by inducing apical bleeding. After the formation of the blood clot, MTA® was inserted and following a period of 24 hours the tooth was restored with glass ionomer and composite resin, and the patient was followed up every six months, for two years. After this period, there was no perceptible mobility in both teeth, and root formation was reached with apical closure in tooth 21. The authors demonstrated that the revascularization technique resulted in better healing and apical maturation.

Still in 2012, Lenzi and Trope [21] described the treatment of two non-vital central incisors with incomplete rhyzogenesis due to trauma. The 8-year-old male patient had coronary fracture in tooth 11 and 21. After coronary opening, rubber dam placement and determination of the working length, copious irrigation was firstly accomplished with 2.5% sodium hypochlorite, and then a new irrigation took place by using antibiotic paste diluted in saline solution. The roots were dried with paper points and filled with the antibiotic past, followed by coronary sealing with glass ionomer® (Vitro Molar; DFL, Brazil). After 35 days the teeth were anesthetized, and the canals were accessed and irrigated with sterile saline solution, and a small bleeding was stimulated. After the clot formation, a mineral trioxide aggregate (MTA Angelus; Angelus, Londrina, Brazil) barrier was made and the tooth was sealed with photo-polymerized bonded resin restoration (3M ESPE, São Paulo, Brazil). At the 4-month follow-up examination, the patient was asymptomatic and the periapical radiographs showed a slight indication that the walls were thickening in the upper right maxillary incisor. At the 11-month follow-up, the patient was still asymptomatic and the revitalization of the root canal was performed since the upper right maxillary incisor showed a distinct thickening of the dentinal walls and closure of the apex of the root, while the upper left maxillary incisor displayed evidence of revitalization. Finally, At the 21-month follow up the patient remained asymptomatic and the successful revitalization of the upper right maxillary incisor was accomplished while the upper left maxillary incisor had not revitalized, but the radiopaque hard-tissue barrier at the apex of it was more distinct. The authors conclude that the complete understanding for the criteria for predictable revitalization and apexification is still lacking.

Kim *et al*. [22] reported three cases, the first being a lower second premolar, in which the 12-year-old patient reported severe pain during chewing. After clinical and radiographic examination, he was diagnosed with pulp necrosis and symptomatic apical periodontitis. Under anesthesia, the root canal was accessed and irrigated, filled with a creamy paste mixture of metronidazole (Samil Pharm, Seoul, Korea), ciprofloxacin (Sinil Pharm, Seoul, Korea) and cefaclor (Myungin Pharm, Seoul, Korea) in sterile saline was applied with the aid of a lentulo-spiral and tapped down into the canal with the blunt end of sterile paper points. The tooth was then temporarily restored using Caviton (GC, Aichi, Japan).

After two weeks, the tooth remained asymptomatic and Caviton was removed under rubber dam isolation and the mixture of antibiotics was completely removed with 3% sodium hypochlorite and sterile saline. Then with the aid of a No. 10 K-file, bleeding was induced; and the blood clot was formed in the root canal around 15 minutes after the stimulation, followed by the application of MTA® (Dentsply Tulsa Dental, Johnson City, TN, USA) directly over the clot followed by a moist cotton pellet. A six-week follow-up showed that the periapical radiolucency had diminished, and the 24-month follow-up finally displayed a completely closed root apex with no periapical pathosis detected. In the second case, a 10- year-old- patient with moderate to severe pain and swelling in the mandibular left second molar was treated. The same methodology as the first case was applied and 42 months later the patient was asymptomatic and without apical periodontitis.

The third case was also accomplished in a young patient as it was in case 2. Following six months of the mandibular left second premolar revascularization, a slight swelling was observed on its buccal surface. During clinical examination, the tooth showed sensitivity to vertical and negative response to percussion tests. Following the radiographic examination, an apical radiolucent image was detected, as well as an incomplete root formation. After complete isolation and anesthesia, the root canal was accessed and there was evident purulent secretion. Irrigation with 20ml of 3% sodium hypochlorite, no more spontaneous drainage existed and the root canal was then filled with a creamy paste as in case 1. The tooth was temporarily restored with Caviton®. In the second session, there was regression of the edema, followed by the removal of Caviton and all the creamy paste. Apical bleeding was then stimulated, and the filling of the root canal with MTA® was made followed by a temporary sealer. Two weeks later, the patient was asymptomatic and the root canal was obturated and the tooth restored with composite resin (Z250; 3M ESPE). After two months, the apical lesions regressed, and 42 months later an increase in the root thickness was observed. The authors concluded that long term forecasts are positive for the revascularization of necrotic teeth with incomplete root formation.

Dens invaginatus is a rare odontogenic formation which happens before the biological mineralization occurs. Treatment options for dens invaginatus include preventive sealing or the filling of the invagination, endodontic treatment, endodontic surgery, intentional reimplant or extraction. Yang *et al*. [23] reported a case of a 11-year-old male patient, who searched for endodontic treatment of the maxillary left lateral incisor referring pain during mastication for the previous two days. The tooth was sensitive to percussion and palpation, low mobility (grade 2) with indication of apical lesion rather than periodontal. The periapical X-ray and cone beam computed tomography (CT) scan revealed type II invagination, which extended from the crown to the middle root. The tooth had pulp necrosis and was isolated under local anesthetic, followed by coronary access with no signs of exudation. Soon after, a second root canal was found. Before instrumentation, the canal was irrigated with 30ml of 5.25% NaOCl, and right after it was filled with a triple antibiotic creamy paste (metronidazole, ciprofloxacin and minocicline) and delivered with a lentulo spiral instrument. The tooth was then temporized with Cavit G (3M ESPE, St Paul, MN). At the following appointment, the patient was partially relieved from the symptoms. After 4 consecutive weeks of medication and irrigation, the patient was completely asymptomatic; there was no sensitivity to percussion or palpation, and no edema. The obturation of the invagination took place with GuttaFlow® (Coltène/Whaledent, Langenau, Germany), and the radiographs showed that it was thoroughly radiopaque. Only after irrigation with 2.5% NaOCl and normal saline and drying the canal with paper points was the apical bleeding induction made with the aid of a #30 K-file, but unsucessfully there was no bleeding at all. After 10 minutes without any sign of bleeding, the root canal access was sealed with glass ionomer cement (Fuji Corporation, Osaka, Japan), followed by composite resin (Filtek Z250; 3M ESPE). Periodical R-ray examinations made every month showed that the periapical radiolucency progressively regressed. Two years later the patient remained asymptomatic, and a periapical X-ray and CT scan revealed that the radiolucent periapical lesion was completely healed, with apical closure and thickening of the root canal walls. The authors conclude that pulp revascularization is an effective new treatment protocol for immature permanent teeth with periapical periodontitis.

The study of Forghani *et al*. [24], in 2013, described a case of a 9-year-old patient in pain when chewing, with localized swelling in the upper anterior region of the maxilla, with history of prior impact trauma three months before the dental appointment. On clinical examination, coronary fractures on both maxillary upper central incisors were detected. The upper right central incisor had a large pulp exposure, sensitivity to palpation and percussion and swelling in the buccal mucosa; and responded negatively to thermal test, being diagnosed as pulp necrosis with an acute periapical abscess. The upper left central incisor had a pinpoint pulpal exposure and no sensitivity to vertical percussion, being diagnosed as irreversible pulpitis. Radiographically, both fractured teeth had immature apices, and a radiolucent periapical lesion around the apex of the right central incisor. After anesthesia with 2% Lidocaine and 1: 800,000 epinephrine (Xylocaine 2%, Dentsply, Addlestone, UK), tooth 21 was accessed and pulpotomy was made. After irrigation with sterile saline, hemostasis was stimulated by a cotton pellet soaked in 5%sodium hypochlorite solution and White Mineral Trioxide Aggregates (ProRoot MTA, Dentsply, Tulsa, OK, USA) powder was mixed with distilled water and placed softly over the exposed clot-free pulpal wound. In the following day, the teeth were restored permanently with composite resin (Filtek Z350, 3M ESPE, St. Paul, MN, USA). The patient was followed up for 6, 12 and 18 months following the treatment, and was asymptomatic in those periods. Radiographically, both teeth showed increased roots and apical closure lengths, although tooth 21 had better results. The authors then believe that revascularization is a suitable treatment for necrotic teeth with incomplete root formation, with apical periodontitis, but suggest further clinical trials in order to prepare a uniform approach strategy.

Jadhay *et al*. [25] evaluated and compared apexification induced by revascularization with and without platelet-rich plasma (PRP) in non-vital immature permanent anterior teeth in 2013. In the first case, the patient was a healthy 10-year-old boy, who was sent for evaluation of the upper anterior teeth fractured subsequent to a fall 3 years prior. Endodontic treatment was initiated by a clinical dentist, but was not concluded. Radiographic examination showed open apexes with incomplete root formation, with thin dentin walls, and when the radiographic data were added to clinical observation, an acute periapical abscess was diagnosed. The intraoral examination showed coronary fracture and edema in the upper central incisors. The teeth were isolated with rubber dam and re-accessed with a round diamond and an endo-Z bur (Dentsply Maillefer, Tulsa, OK). A purulent discharge was then observed and the canals were irrigated with saline, followed by the establishment of the working length with the aid of paper points (Dentsply Maillefer, Tulsa, OK) and confirmed with a large file in the canals and periapical X-rays. Minimal mechanical instrumentation with a 60H-file (Dentsply Maillefer, Tulsa, OK) and irrigation with 20 mL 2.5% sodium hypochlorite (NaOCl, Cmident, India) were performed. The canals were dried with paper points and a triple antibiotic paste, mentioned by Hoshino *et al*. 1996, was applied with a sterile number 30 hand lentulo spiral (Dentsply Maillefer, Tulsa, OK). The temporary restoration was made followed by intermediate restorative material (Caulk Dentsply, Milford, DE). Local anesthetic solution without adrenaline (LOX 2% Neon Lab, India) was infiltrated around the apices of both the central incisors and a sterile endodontic file, endowed with a rubber stopper was set at 2 mm beyond the established working length. The file was then pushed past the confines of the canal into the periapical tissue in order to cause a frank bleeding. Afterwards, a dry cotton pellet was inserted 3-4 mm into the canal and held there for 5-7 min to allow blood clot formation in the apical third. This protocol was followed for both teeth involved.platelet-rich plasma (PRP) was prepared in a table top laboratory centrifugation machine (Remi model no. - R-8C, India) separate the PRP from the platelet-poor plasma (PPP). Freshly prepared PRP, soaked on a 1 $mm^2 \times 1$ mm^2 sterile collagen sponge (Metrogene, Septodont, France) was introduced into the root canal of maxillary left central incisor with cotton pliers and carried to the middle third with a size 30 finger plugger (Sybronendo, CA, USA). Access openings were restored with resin modified glass ionomer cement (Photac-Fill, 3MESPE, Minnesota). Standardized intraoral periapical baseline and subsequent follow-up radiographs at 6 and 12 months were taken with a Rinn positioning device (Dentsply, Elgin, IL).

In the second case, a 23-year-old patient was referred with complaints of prolonged pain in the upper anterior teeth. The patient had a history of trauma 15 years before, which went on untreated. The intraoral examination revealed discoloration on both upper central incisors and the presence of a fistula on the palate in the upper left lateral incisor region. Radiographic examination showed a radiolucent image in both upper central incisors and upper left lateral incisor. Both central incisors had open apexes and thin dentin walls, and the diagnosis was pulp necrosis with chronic apical abscess for all the teeth involved. The upper left lateral incisor was treated by the conventional endodontic therapy. Revascularization with and without PRP was randomly induced on the upper central incisors, left and right, respectively. Final aesthetic rehabilitation was held with porcelain crowns.

In the third case, a 13-year-old patient was sent for endodontic evaluation referring continuous pain in the previous three months in the upper anterior teeth. During clinical and radiographic examination, edema and a radiolucent image were detected in both teeth, which had open apexes and thin dentinal walls. The diagnosis was chronic abscess for both teeth. After revascularization and infection control, the teeth were randomly treated with and without PRP. Radiographic examinations were accomplished after 6 and 12 months, and all three teeth involved were asymptomatic.

The authors conclude that revascularization is an effective method for the induction of maturation in non-vital teeth with incomplete root formation, and also that the treatments with PRP can potentially improve and accelerate the desired biological result of such regenerative technique. The kinds of intracanal medication used by the authors in this review can be observed in Table **1**.

Table 1. Intracanal substances used for cell stimulation and infection control.

Authors	Intracanal medication	Paste components
Aggarwal	Calcium Hydroxide Paste Paste	Calcium Metronidazole+ciprofloxacin+ mynociclin
Jadhay	Paste platelet-rich plasma (PRP)	Metronidazole+ciprofloxacin+ mynociclin
Kim	Paste	Metronidazole+ciprofloxacin+cefaclor Metronidazole+ciprofloxacin+mynociclin
Kvinnsland	Paste	Calcium hydroxide + IRM
Lenzi and Trope	Paste	Metronidazole+ciprofloxacin+mynociclin + saline
Nevins	Gel	Collagen-calcium phosphate
Nygaard and Ostby	Paste	Koplerca N-O
Pramila and Muthu	Paste	Metronidazole+ciprofloxacin+ mynociclin
Rule and Winter	Paste	Polymix B sulfate+neomycin sulfate+bacitracin+nystatin

THE ROLE OF HUMAN PULP DERIVED STEM CELLS AND OTHERS

Researchers have, since 2008, focused on the potential applications of Human Pulp Derived Stem Cells (HPDSCs) for regenerative procedures, in order to produce tissues *in vitro* that can be inserted in the human body, as well as on the usage of preexistent cells in certain tissues that can be professionally stimulated or in naturally occurring sites of the human body. The periapex is a region enriched with different sorts of traditional cells interacting with stem cells correspondent to the varied tissues present therein. Stem cells like Periodontal Ligament Stem Cells (PDLSCs), Stem cells from Human Exfoliated Deciduous Teeth (SHEDs), Dental Pulp Stem Cells (DPSCs), Stem Cells from the Apical Papilla (SCAPs) and Dental Follicle Stem Cells seem to play important roles in the healing process, especially for Regenerative Endodontic Procedures (REPs), which according to some authors seem to promote guided endodontic repair rather than a true regeneration of physiological-like tissue; due to the fact that such cells differentiate under stimulation [26, 27]. There have been a number of articles describing the possibilities of differentiation that HDPSCs may display, including them being precursors of main cells types like osteoblasts, cementoblasts and fibroblasts among many others.

DISCUSSION

Infection control seems to be crucial for the success of pulp revascularization. In the majority of the experiments described in this review, the three antibiotic paste (TAP) consisting of metronidazole, ciprofloxacin and mynociclin was the medication most used. Metronidazole is an antiprotozoal, antibacterial and antihelminthic nitroimidazole agent with special interest for endodontics for disrupting energy metabolism of anaerobes by hindering the replication, transcription and repair process of their DNA. Anaerobic presence in the root canals implies in more resistant infections, and therefore the association with ciprofloxacin, which has activity against a wide range of gram-negative and gram-positive bacteria added more efficacies to the paste. Up to this moment, there is no known cross-resistance between ciprofloxacin related in the endodontic microflora. Minocycline is a broad-spectrum tetracycline antibiotic, with a broader spectrum than the other members of the group. It is a bacteriostatic antibiotic, classified as a long-acting type. This paste seems to help control bacterial infection within the root canal space, more specifically in the periapex location.

Regenerative procedures constitute an emerging field in the health sciences and particularly in odontology, since the steps can be followed up with the aid of computer cone beam tomography or traditional and digital periapical radiographs, because the root formation process involves hard tissue deposition and can be followed, measured and compared to previous examinations. In the near past, there seemed to be a resistance of some authors as for trying revascularization in infected, nonvital, immature teeth, simply because this possibility although foreseen by some researchers, was considered uncertain because of the myth that it would be too risky trying to revascularize an infected root canal. Therefore, the traditional treatment of inducing apexification with calcium hydroxide and mineral trioxide aggregate or with surgical endodontic procedures to seal wide-open apex teeth was always chosen as the first line of endodontic approach. Although the interactions with HDPSCs is not yet very clear [28, 29], due to the complexity and lack of information, traditional pulp cells that survived infection may proliferate under the influence of Hertwig's epithelial root sheath even during the inflammation process; giving rise to odontoblasts that are able to populate atubular dentin at the apical end, stimulating apexogenesis [30 - 32]. The inflammation process requires good blood supply present in the periapex, in order to recruit cells for the defense against pathogens. Therefore, chemotactic factors are liberated, known as cytosines and interleukins. The large cytokine family includes the Interleukins (IL), Interferon (IFN), Tumor Necrosis Factor (TNF), Coloning-stimulating Factor (CSF), Chemokines (CKs), and Growth Factor (GF). Together, or alternately, they stimulate inflammation in some situations while in others simply restrain it, modulating the process. Some of such traditional cells are the Mast cells, which, when stimulated by etiological factors such as bacterial neurotoxins, increase in number and undergo degranulation, causing inflammatory and vascular changes [33, 34]. Nevertheless, for promoting healing and enhancing revascularization, root canal infection must be removed.

Another mechanism for root development can be attributed to stem cells from the apical papilla (SCAP) or the bone marrow stem cells present in the alveolar bone. Once the instrumentation is made beyond the limits of the root canal into the periapex in order to induce bleeding, there may be the transportation of Mesenchymal Stem Cells (MSCs) from the bone into the canal lumen, giving rise to bone or dentin-like tissues *in vivo* [35, 36].

From the studies mentioned in this literature review, it seems that the use of antibiotic creamy pastes has led to satisfactory results in pulp revascularization, as it helped controlling infection which led to the increase in root thickness enhancing apical closure, as demonstrated by the studies of Trope [14]; Pramila and Muthu [19]; Lenzi and Trope [21]; Kim [22]; and Yang [23]. Kvinnsland [15], who used calcium hydroxide in revascularization, found the same satisfactory results. Forghani [24] used MTA only as a medication for revascularization, and also obtained satisfactory results for treating teeth with incomplete root formation.

In comparative studies evaluating the use of topic antibiotics and calcium hydroxide, used in pulp revascularization, Aggarwal [20] reinforced that among all the elements used, the antibiotic creamy paste resulted in better apical healing when compared to calcium hydroxide.

The main reason for such satisfactory results is that revascularization is not possible in the presence of necrotic pulp tissue occupying the root canal space. The toxins liberated from bacteria in their course for decomposing pulp tissue irritate the periapical area, while stimulate immune responses which, in the attempt of eliminating the pathogens, also destroy soft and hard tissues. After infection has been controlled, and the root canal space is disinfected, capillar neoformation takes place in order to reestablish blood flow, which now will nourish cells with the appropriate nutrients necessary for homeostasis. Among the cells involved in the process of revascularization, Human Pulp-Derived Stem Cells (HPDSCs) seem to play important roles, since they are able to differentiate into the main cells responsible for root formation and angiogenesis. Self renewal, high proliferative activity associated with the ability of forming colony formation units (CFU) are some of the characteristics that make these precious cells play important roles in helping closing the open apexes. They are able to differentiate into osteoblasts, odontoblasts, chondrocytes and myocytes, and also fibroblasts. Among the most important HDPSCs; DPSCs, PDLSCs and SCAPs are interestingly connected to the periapical region. Together, associated with traditional immune cells like lymphocytes and fibroblasts, the apex begins to heal, enhancing closure [37].

CONCLUSION

Although pulp revascularization is a recent therapy of regenerative endodontic procedures, it seems to be effective for immature teeth since it allows root formation in a relatively simple technique, and to enhance prognosis for the treated teeth. Nevertheless, more studies are necessary to evaluate its long-term efficacy and new approaches.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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