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Evolution of regenerative endodontic in young permanent teeth: a literature review

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Abstract

Objective: to compare different protocols available for regenerative endodontic. Methodology: articles published between 2001 and 2021 were searched on the database PubMed using the following keywords: "pulp revascularization" or "pulp revitalization" or "apexification". Inclusion criteria were studies on humans, patients with immature permanent teeth with necrosis or irreversible pulpitis, a follow-up of at least 12 months, clearly described protocols and in English language. Results: The search produced 944 results. After applying the inclusion and exclusion criteria, only 35 were selected. 11 articles were added following a bibliographic review of the selected articles and a total of 46 cases were included. The success rate outcome for regenerative endodontic was of 80% observed in a total of 278 teeth. During irrigation, NaOCL was used in most of the cases with concentration varying from 0.5% to 6% and TAP has been the most used intracanal medication (32.7%). Periapical healing and resolution of the symptoms were observed in 95.7%, while apexogenesis was observed in 80.2%. Increase in root length reach 60%, root thickness 64.8% and apical closure 61,9%. Pulp vitality was found in 7.2% of the cases and discoloration in 29.5%. Conclusion: Regenerative endodontic showed signs of apexogenesis in most cases, and a pronounced increase in root thickness, root length and closure of the apex. Due to a high number of protocols and their heterogenicity, the superiority of one over the others cannot be established. More randomized clinical trials are needed to determine the most effective protocol.

Keywords: Regenerative Endodontics; Apexification; Dental Pulp Cavity; Disinfection.

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Introduction

Immature permanent teeth are defined as newly formed teeth that have not completed their apex/root formation and thus possess an open apex[1]. Dental trauma occurs in children undergoing cranioskeletal development at a rate that vary between 2,6% to 35%[2]. These traumas and pathologies, together with developmental anomalies such as dens evaginatus and dens invaginatus usually provoke pulpar pathologies that if left untreated, will lead to subsequent periapical pathologies[3]. It could affect the Hertwig's Epithelial Root Sheath (HERS) which is responsible for the differentiation of odontoblast from the apical papilla as well as determining the final shape of the root[3]. Apical papilla is in short, a new terminology for the dental papilla located at the apex of growing immature permanent tooth[4]. It was recently discovered that it contains new types of stem cells apart from the dental pulp stem cells (DPSCs) named stem cells from the apical papilla (SCAP)[4]. DPSC were considered to be the origin of odontoblast although it seems that SCAP are the ones that differentiate into the primary odontoblast responsible for root dentin formation while DPSC differentiate into replacement odontoblast[4].

Immature permanent teeth were treated by apexification before regenerative endodontic appears, but the outcome is limited as it's only intended to create an apical calcified barrier through calcium hydroxide or the apex closure through mineral trioxide aggregate (MTA)[5] or Biodentine[6].

Regenerative endodontic is defined as a "biologically based procedures designed to replace damaged tooth structures, including dentin and root structures, as well as cells of the pulp-dentin complex"[7]. It relies on the potential that mesenchymal stem cells located at the apical papilla will differentiate into odontoblast upon induction from growth factors once they invade the root canal after induced bleeding at the apex[2]. The AAE (American

Association of Endodontists) has defined three objectives for this procedure to be successful[5]:

- Primary: Elimination of symptomology and evidence of bone healing.
- Secondary: Increase in root wall thickness and/or in increase root length.
- Tertiary: Positive response to vitality test.

While the primary objective is common to every endodontic treatment and indispensable (being fulfilled in more than 90% of cases in regenerative endodontic procedures (REP)[2]; the second one, being the real added value of REP, is a desired outcome which shows variabilities in distinctive clinical cases. In a review of clinical cases from Chen *et al.*[8] (2015), increased root length was observed in 76,2%, increased root width in 79,2% and apical closure in 55,4% of cases. The third objective being the ultimate goal and the leading objective, in the current research has been witnessed around 50% to 60% of the cases according to two reviews[5]. However, without histologic analysis it's not possible to confirm that the formed tissue was true pulp tissue and not just vital tissue that is known to be innervated[5].

While regenerative endodontic is an easily duplicated procedure, it remains unpredictable as it is subjected to many factors: operator variability, duration and extension of the pathology, stage of the root development, size of apical diameter and finally the type and concentration of irrigant, antibiotics or disinfectants being used.

Mechanical debridement is one of the keys to success of conventional endodontic treatment but in regenerative endodontic procedure, due to the intrinsic characteristic of the tooth, it is advisable to perform little to no mechanical debridement to preserve the already thin dentinal wall and impede any fracture risks[9].

Irrigation with sodium hypochlorite (NaOCL) is the most widely used[10] as it holds effective bactericidal capacity, lubrication capacity and tissue dissolution capacity, property that does not share chlorhexidine (CHX)[11]. The main roles of chemical irrigants are control of bacteria inside the root canals (being the most essential one), as well as the impediment of debris compaction and extrusion of bacteria at the apex. It was established that organic and inorganic compound of the dentin in suspension in a solution could alter the bactericidal efficiency of the irrigant[11]; thus, the AAE in the protocol for REP recommended a copious irrigation of 20 mL of sodium hypochlorite of the canal to renew the irrigant and remove the organic and inorganic compound present in the root canal[9]. Sodium hypochlorite has proteolytic activity over dentin collagen matrix when used in high concentration[12] and has shown to affect those structural characteristics by decreasing elastic modulus and flexural strength significantly when used in high concentration (concentration of 5.25% compared to one of 0.5%)[13]. On the other end, previous studies exposed the fact that reduction of intracanal bacteria was not greater at a 5% concentration in comparison to one of 0.5%[12]. In addition to the previously established key factors in successful endodontic outcome (bacterial control and structural strength maintenance) new key factors in the achievement of REP have emerged: maintenance of viable stem cells together with growth factors availability. Following the discovery made in 2011, it was detected that after evoked bleeding, numerous mesenchymal stem cells were delivered into the root canal[10].

Considering the central role that play stem cells in the success of regenerative endodontic, irrigation should not only be guided by the need of bacterial control but also by its possible deleterious effects on stem cells. It has been demonstrated that NaOCL, at a concentration of 0.5%, 1.5% and 3%, decreased the availability of stem cells from the apical papilla (SCAP) by around 37% whereas at a concentration of 6%, the reduction was very significant around 85%[14]. This deleterious effect of NaOCL was partially reversed after using 17% EDTA (Ethylenediaminetetraacetic acid)[14]. Another mineralization precursor of odontoblast, dentin sialophosphoprotein (DSPP) is affected by NaOCL depending on its concentration. At a concentration of 1.5%, NaOCL does not affected DSPP gene expression, whereas at 3% reduces DSPP expression by half and completely erases its expression when NaOCL is used at 6%[14]. Those effects were reversed, one completely and the other by half (respectively) after the use of 17% EDTA. NaOCL has demonstrated to have long lasting effect on dentin that couldn't be attributed to the presence of remaining irrigant in the dentinal tubule. After its deactivation with 5% sodium thiosulfate the negative effects of NaOCL were not reversed[10]. Those effects include denaturation of dentin-derived growth factors together with a decrease in dental pulp stem cells (DPSCs) attachment on dentin[14]. DPSC exhibited *in vivo* higher proliferation rate and higher number of odontoblast-like marker when placed in a tooth-sliced model in comparison to DPSCs placed in a scaffold only[15]. It could be established that morphogens and more precisely growth factors, play a key role in dental stem cells survival, proliferation and differentiation[15].

EDTA has shown to solubilize growth factors from dentin and thus to increase their bioavailability[14]. It also promoted stem cells differentiation and attachment[10]. A balance should be found between the leading concepts around bacterial control and the emerging ones around stem cells and growth factor survival. Therefore, the actual recommendations for chemical irrigation are the use of 1.5% NaOCL followed by 17% EDTA.

Another essential point in regenerative endodontic is the intracanal medication used. The first one used was the double antibiotic paste (DAP) by Iwaya *et al.*[16] (2001) composed of metronidazole and ciprofloxacin. Later minocycline was added to what would soon become the triple antibiotic paste (TAP). This modification was created because of previous studies showed the efficacy of the TAP to eliminate bacteria *in vitro* with a concentration of 100 µg/ml[10]. *In vivo* study on dog teeth also revealed the efficacy of the TAP, being effective in eliminating cultivable bacteria in 70% of the treated teeth and where the remaining bacteria were only present in a very small amount[10]. It was then modified again: minocycline, which is associated with discoloration[18] has been replaced by cefaclor[17]. In many case reports, clinicians focused on the consistency of the TAP and not on the concentration. They wanted to achieve a creamy texture by mixing it with water, saline or propylene glycol, favouring its handling and delivery into the canal[10]. But, at this described consistency, the concentration of antibiotic is around 1.000 mg/mL which is known to have detrimental effect over survival of SCAPs[19]. In study of Ruparel *et al.*[19] (2012) the effect of different concentrations of antibiotics commonly used in regenerative endodontic was analysed; it was found that all the antibiotics studied (DAP, TAP, modified TAP with cefaclor and Augmentin) had a detrimental effect over SCAP survival when used at a concentration of 1 mg/mL, 10 mg/mL, 100 mg/mL and 1.000 mg/mL. This detrimental effect followed a concentration dependent pattern. The use of antibiotics at a concentration of 0.1 mg/mL and 0.01 mg/mL is safe over SCAP survivals and effective against endodontics bacteria. In their comprehensive review, Kim *et al.*[5] (2018) reminds that Augmentin, despite being effective in bacterial destruction, only affects bacteria's cells wall (which is absent in human cells) thus it is not supposed to be harmful on human cells, but according to the study of Ruparel *et al.*[19] (2012), at a certain concentration, Augmentin decreases survival rate of SCAP. Antibiotics exhibited several problems: their potential to induce bacterial resistance, the possible systemic allergic reaction, the cytotoxic effects on cells and the problem of removability[20]. Following different common irrigation methods, it was revealed that more than 80% of TAP was still present in the canals whereas more than 85% of calcium hydroxide was removed[10]. Despite its effect on SCAP, TAP have a negative impact on growth factors released from dentin, while calcium hydroxide positively influences their release[20]. Calcium hydroxide on the other hand, doesn't affect survival of SCAPs[19] and exhibits bactericidal properties[21]. It also promotes the release of bioactive molecules from the dentine matrix which possess the capacity to support reparative dentinogenesis[22]. However, Yassen *et al.*[23] (2013) showed that the use of calcium hydroxide, DAP and TAP for a long period of time (3 months) had a significant impact on fracture toughness and reduced it by 30%, 21% and 19% respectively. TAP and DAP additionally, significantly decrease the root microhardness. Thus, the additional effect of EDTA irrigation on demineralization should be considered when selecting the intracanal medication. At the time of intracanal medication election, the clinician should consider: its efficiency to create a bacteria-free environment, it's possible effect on dentine matrix (structural strength, growth factors release), its consequence on SCAP and the potential for adverse reactions.

Regenerative endodontic procedures rely on stem cells' potential[5]. They have the ability to differentiate into different phenotypes (odontoblasts, neural progenitors, osteoblasts, endothelial cells) upon induction from environmental stimuli such as, hypoxia, extracellular matrix, growths factors or other conditions[3]. To achieve the desired result in regenerative endodontic, namely the recovery of a functional dentinal-pulp complex, three major principles of tissues engineering are applied[17]. They enclose an adequate source of progenitor/stem cells, viable growth factors capable of inducing differentiation of stem cells and a 3D scaffold that regulate stem cells differentiation through local release of growth factors[3]. The main source of stem cells in REP is the periapical tissue[17]. Following laceration of the apical papilla, the influx of blood inside the root canal contained a concentration of markers specifics to undifferentiated mesenchymal stem cells 400 to 600 times superior[3]. Following this influx, stem cells should be guided by growth factors which regulate survival, migration, proliferation and differentiation of cells[5]. Mineralized dentin contains several biological molecules such as glycosaminoglycans, non-collagenous proteins and growth factors which are solubilized during dentin conditioning by EDTA. They could orientate the behaviours of stem cells[5].

Different essential families of growth factors are needed to achieve a functional dentinal-pulp complex. Transforming growth factors β (TGF- β) are associated with cell proliferation, extracellular matrix synthesis, odontoblastic differentiation and dentinogenesis. They also have a chemotactic effect on dental pulp stem cells (*in vitro*) and play a role in immune response of the pulp. Platelet derived growth factors (PDGF) act on angiogenesis and promote cell differentiation, dentin matrix synthesis, odontoblastic differentiation and dentinogenesis. Bone morphogenic protein (BMP) act on odontoblastic differentiation and dentinogenesis. Vascular endothelial growth factors (VEGF) promote odontoblastic differentiation and cell proliferation and act on angiogenesis. Fibroblast Growth factors (FGF2) promote chemotaxis, angiogenesis but also cell proliferation and dentinogenesis. Insulin-like growth factors (IGF) act on cell proliferation and odontoblastic differentiation. Nerve growth factors (NGF) encourage cell differentiation and angiogenesis[24].

The role of SCAP in the root development hasn't been clearly established yet, but still it has been enlightened by previous studies. It was observed *in vivo* that in teeth where the pulp tissue was removed, and the apical papilla retained (following trauma and pulp exposure) a continuation of root tip formation could be appreciated[4]. Further histological examinations are needed to confirm if the formed tissue is dentin or cementum. Another interesting study conducted in minipigs, exhibited the discontinuation of root formation when apical papilla was removed at early stage of tooth development[4]. Even though the discontinuation in formation could be due to a damaged HERS during surgical removal of SCAP, those results are promising in showing the role of SCAP.

To guide the influx of stem cells, a scaffold is needed. It's one of the main elements of REP and consists of a three-dimensional structure that allows cell binding, cell localization and which provides local release of growth factors[25] through platelet activation[26]. It should be biodegradable and simulate the extracellular matrix[5]. Blood clot (BC) is the most widely used scaffold, and besides being cheap, it does not require manipulation by the clinicians and provides the entry of numerous cells into the root canal. However, the types of cells and their concentration couldn't be selected, making the outcome unpredictable[25]. Another issue that could appear would be the impossibility of inducing bleeding into the root canals, maybe because of an extended periapical destruction. This concern might be solved either by postponing the procedure until recovery from the periapical tissue[5] or by going through an apexification procedure in single rooted teeth (as for multi-rooted teeth, bleeding can be harvested from the other canals)[25]. Autologous platelet concentrates have been used as scaffolds (either alone or together with induced bleeding) because of their high concentration of platelets and their possible improvement of regenerative endodontic outcomes[27]. Platelet rich plasma (PRP) and platelet rich fibrin (PRF) are the most used. Platelet pelet (PP) have been tried for the first time in REP by Ulusoy *et al.*[27] (2019). Even though their use was associated with positive clinical and radiological outcomes, following histological analysis, no pulp like-tissue or odontoblastic cells were found[25]. Additionally, no differences were found in terms of root maturation between PRP, PRF, PP and BC[27]. Thus because of the high cost of autologous platelet concentrate and their small benefice over BC, their use isn't recommended[25]. The development of an artificial scaffold that would contain a cocktail of different growth factors at precise concentrations and with angiogenic, proliferative, differentiative and chemotactic properties specific of the dentinal pulp complex should be investigated.

Even though promising, regenerative endodontic face some adverse effects such as discoloration that is one of the most known adverse effects. It affects around 40% of teeth following REP[5]. It is usually associated with TAP that contained minocycline[9]. An alternative to minocycline involves the use of DAP or modified TAP (with cefaclor)[18]. Capping material such as MTA (grey or white) and calcium hydroxide are known to produce discoloration as well. Therefore, when working on aesthetic anterior sectors, the choice of material is of particular importance and Biodentine could be used to avoid crown discoloration[5]. Bacterial reinfection, because of improper canal disinfection or bacterial leakage may also happened and lead to failure of the procedure. Another major issue are fractures, which have been observed in some cases. Ultimately, root canal calcification following regenerative endodontic was found in 62.1% of the cases[5]. Its prevalence might be increased when using blood clot as a scaffold, as blood clots have been associated with a progressive root canal obliteration independently of the intracanal medication used (TAP or calcium hydroxide)[27]. Therefore, AAE does not recommend the use of regenerative endodontic if the pulp space is needed for a post/core as final restoration[28]. Moreover, the calcification of the canal could be challenging if the tooth suffers reinfection and need to undergo root canal treatment[18]. The protocol given by the AAE is presented in Table 1.

Objectives

At the light of the several possible approaches of regenerative endodontic, through different protocols and the observation that they all possibly could be successful, the main objective of this work is to compare the different protocols available. To support this work and answer to this objective, two secondary objectives have been established: compare the short and long-term outcomes of each protocol through the review of clinical cases and the determination of the advantages and disadvantages of each protocol.

TABLE 1 · AAE Clinical consideration for a regenerative procedure[28]

First appointment:
» Local anesthesia, dental dam isolation and access.
» Copious, gentle irrigation with 20ml NaOCL (using an irrigation system that minimize the possibilities of extrusions). » Irrigation with 1.5%-3% min (20mL/canal, 5 min) NaOCL. » Then irrigation with saline or EDTA (20 mL/canal, 5 min), (needle positioned 1 mm from root end).
» Dry canals with paper points.
» Place calcium hydroxide or triple antibiotic paste (TAP). If using the TAP: • consider sealing pulp chamber with dentin bonding agent. • mix 1:1:1 ciprofloxacin: metronidazole: minocycline to a final concentration of 1-5 mg/ml.
» Deliver into canal system via syringe • If triple antibiotic is used, ensure that it remains below CEJ (minimize crown staining).
» Seal with 3-4mm of a temporary restorative material (e.g. Cavit, IRM, glass-ionomer…).
Second appointment: 1-4 weeks after 1st visit
» If there are signs/symptoms of persistent infection: • consider additional treatment time with antimicrobial, or alternative antimicrobial.
» Anesthesia with 3% mepivacaine without vasoconstrictor, dental dam isolation.
» Copious, gentle irrigation with 20ml of 17% EDTA.
» Dry with paper points.
» Create bleeding into canal system by over-instrumenting (pre-curved endo file, endo explorer, 2 mm past the apical foramen) • The goal is to have the entire canal filled with blood up the cemento-enamel junction.
» Alternative to blood clot is the use of platelet-rich plasma (PRP), platelet rich fibrin (PRF) or autologous fibrin matrix (AFM).
» Stop bleeding at a level that allows for 3-4 mm of restorative material.
» Place a resorbable matrix (e.g. CollaPlug, Collacote, CollaTape) over the blood clot if necessary and white MTA as capping material.
» A 3–4 mm layer of glass ionomer is flowed gently over the capping material and light-cured for 40 s
» MTA has been associated with discoloration. Alternatives to MTA (such as bioceramics or tricalcium silicate cements should be considered in teeth where there is an esthetic concern.
» Anterior and Premolar teeth: • Consider use of Collatape/Collaplug and restoring with 3mm of a non-staining restorative material followed by bonding a filled composite to the beveled enamel margin.
» Molar teeth or teeth with PFM crown: • Consider use of Collatape/Collaplug and restoring with 3mm of MTA, followed by RMGI, composite or alloy.
Follow-up: (6, 12, 24 months)
» Clinical and Radiographic exam: • No pain, soft tissue swelling or sinus tract (often observed between first and second appointments). • Resolution of apical radiolucency (often observed 6-12 months after treatment) • Increased width of root walls (this is generally observed before apparent increase in root length and often occurs 12-24 months after treatment). • Increased root length. • Vitality test
» Recommended yearly follow-up after the first 2 years
» CBCT is highly recommended for initial evaluation and follow-up visits

Methodology

To conduct the research of scientific articles for the review, articles ranging from 2001 to 2021 on the database PubMed, clinical research on revascularization using the following keywords: "pulp revascularization" or "pulp revitalization" or "apexification" was conducted on July 2021.

Inclusion criteria were studies conducted on humans, patients with immature permanent teeth suffering from necrosis or irreversible pulpitis, a follow-up of at least 12 months in each case, clearly described protocols and in English language.

Exclusion criteria were study on animals, patient with permanent mature teeth, a follow-up of less than 12 months, unclear protocols, studies in languages other than English, insufficient data. Not from the following journals: Journal of Endodontic, International Endodontic Journal, Dental Traumatology, Pediatric Dentistry, Journal of Clinical Pediatric Dentistry.

Results

The search produced 944 results. 282 were removed because the text was not full, 18 were not in English, 35 were eliminated as they were not within the date range established and 225 were not conducted on humans. 152 were excluded as they were not from the selected journals. At this point, 232 articles were eligible but from those only 35 were selected, the rest were discarded as they did not have a sufficient follow-up, or the clinical protocol was unclear, or the subject was not about regenerative endodontic. 11 articles were added following a bibliographic review of the selected articles and a total of 46 cases were included.

The following information were collected: sample size, patient age, pathology of the patient (pulp state, periapical state, aetiology), mechanical debridement, irrigation (type of irrigant and concentration), intracanal medication (type and concentration), final irrigation (type and concentration), evoked bleeding, additional scaffolds, follow-up, outcomes (resolution of the periapical pathology, increase in root length, increase in root width, apical closure, failure), adverse effects (discoloration or others) and response to vitality testing.

The tooth was considered as whole, thus for multirooted teeth if a canal presents a characteristic and/or pathology, it was recorded as if all the root canals were subject to it. In the articles, an indicated "continued root formation and/or physiological root formation" which informed of the outcome was recorded as an increase in length and width. Minimal instrumentation referred to a light instrumentation up to the apical third whether partial instrumentation referred to an instrumentation of the coronal and/or middle third. The case was considered a failure if the tooth did not achieve the primary objective (elimination of the symptoms and the evidence of bone healing) but also if it did not achieve one of the secondary objectives (increase root wall thickness and/or increase root length and/or apical closure) or if it was recorded by the author as failure.

The data of included studies are presented on Table 2 (Pre-treatment), Table 3 (Post-treatment), Table 4 (Demographic and study data) and Table 5 (Follow-up variables).

Discussion

According to this review, 278 teeth were studied. The success rate outcome for regenerative endodontic was of 80%. This means that in those cases, some increased root length, thickness and/or apical closure were detected. However, care must be taken at the time of interpreting the results as the majority of the data was extracted from case reports and/or case series and not only from randomized clinical trials. Thus, this data might be subject to bias publication as authors are more prone to report cases with positive outcomes rather than cases with negative outcomes. Another important point at the time of interpreting the results is the great diversity in protocols and the lack of standardized analysing methods. Although no differences were found between 2-dimensional radiograph and CBCT at the time of assessment in root development[29], the use of standardized analysing methods for each patient and at each appointment is mandatory to reduce the risk of distortion due to angulation which could alter the results.

Mechanical debridement

Regarding instrumentation, one third of the cases were found to have some kind of root canals mechanical debridement which is not in accordance with the clinical consideration for regenerative procedure proposed by the AAE. It is supposed that authors had chosen to use it following the discovery that failure in REP were mainly due to remaining bacteria in the apical third[9]. As the procedure is performed on teeth with thin dentinal wall, the potential benefit on bacterial control might not exceed the risk of weakening the already fragile, fracture prone immature teeth[30]. The instrumentation created a smear layer that will occlude the dentinal tubule and alter the cell migration, proliferation and adhesion[31]. However, in the thirteen cases with a failed outcome[29,32-43], only four used instrumentation[29,35,38,40]. Among the failed cases, EDTA was used in two study[29,42]. EDTA use (a chelator agent) as final irrigation solution for chemical debridement has the potential to remove the smear layer created during the mechanical debridement and also promotes the release of growth-factors entrapped into dentin that will enhance differentiation of stem cells into odontoblast-like cells[14]. This property of EDTA is known by many clinicians but it was only used in 10 of 46 studies which represent 40.7% of the treated teeth. It was used with at a concentration of 17% except in one study which used it at 5%[44] and another one which use it at a concentration of 15%[7].

Chemical debridement

During irrigation, NaOCL was used in the majority of the case, either alone (65.2%) or together with another irrigant (30.4%). Its concentration varied from 0.5% to 6% but in many studies, it was used at concentration higher than 5%. The rationale for using this high concentration is unidentified especially because it was demonstrated that 5% NaOCL did not eliminate more bacteria inside the canal compared to 0.5% NaOCL[12] and that 5.25% NaOCL significantly decreased the elastic modulus of dentin compared to a 0.5% concentration[12]. It was also stated that it is the TAP and not the irrigant that have the greater bactericidal effect[8]. Six of the failed cases used a concentration of 2.5% or 2.6%[32,34,36,40,41,43], four used a concentration of 5.25%[29,27-29], one used either 3% NaOCL or 0.12% chlorhexidine[35] while the rest used a combination of different irrigants[33,42]. Although it was demonstrated that NaOCL had deleterious effects on stem cells survival, especially when used a 6%[14] and if EDTA (that could reverse this deleterious effect) use was sparse; many more cases should have failed as stem cells are believed to be the corner stone in successful regenerative endodontic and that theoretically stem cells should have been damaged. In some cases, chlorhexidine was used either alone or together with NaOCL at a concentration that varied from 0.12% to 2%. While the cytotoxicity of chlorhexidine over stem cells have been verified[45], 11 studies used it in their protocols and apart from 2 cases, all were successful. Among them 4 used EDTA as a final irrigation. Three studies used H_2O_2 together with NaOCL and all were also successful.

Intracanal medication

Regarding intracanal medication, TAP has been the most used (32.7%). It is plebiscite for its powerful antibacterial properties studied *in vitro*[5], but it also raises concern for its potential systemic allergic effect, for the development of bacterial resistant strains[5]. TAP moreover has a harmful effect over stem cell when used at high concentration which is often the case as many authors are concerned about the consistency of the paste and not about the concentration of antibiotics[20]. It also appears that the elimination of the paste is complicated and that antibiotics have a profound effect on dentin[10]. TAP has been associated with discoloration problems due to minocycline[29] which explained that some authors removed it from the formula or that it was replaced by cefaclor, doxycycline or amoxicillin. Seven cases that used TAP exhibited discoloration[26,29,35,39,46-48] including one from

McTigue *et al.*[35] (2013) that change to TAPc after observing discoloration. Three others case that used substitutes of minocycline such as TAPm[6], TAPc[42], TAPa[49] and calcium hydroxide[50] also suffered discoloration. Calcium hydroxide is also recommended by the AAE as intracanal medication due to its antibacterial properties. It appears to have a positive impact over stem cells from the apical papilla[10]. One of its adverse effects which is also common to the antibiotic paste, is its effect on dentin structure[10] but this deleterious effect only appears 2 months after the medication has been placed inside the canal[20].

Evoked bleeding

There is one successful case of regenerative endodontic in which the author decided not to provoke bleeding. The pulp was necrotic but the author used an additional scaffold of PRF[51]. This scaffold rich in growth factor attracted apical stem cells and replaced the need for evoked bleeding[25]. Three other cases failed to induced bleeding inside the canal. One from Petrino *et al.*[33] (2010): bleeding was not achieved during the first attempt due to the use of 2% lidocaine as anesthetic. At the second attempt, mepivacaine was used and bleeding was present but at an insufficient level to allow blood clot formation and subsequent placement of MTA. The desired bleeding was achieved at the third attempt and MTA was placed over the clot. The tooth did not showed sign of apexogenesis. It might be due to the presence of a big apical abscess that formed after a trauma 6 years prior to the consultation and that could have injured the HERS and the stem cells as apical disease of more than 6 months have been suggested to be responsible for the absence of viable apical stem cells[18]. Also, the repeating irrigation with 5.25% NaOCL and 0.12% CHX might have damaged the growth factors presents in the dentin. One from Yang *et al.*[48] (2013): no bleeding could be achieved during the first attempt, the author decided to use a glass ionomer cement as a capping material and to control the evolution over 24 months. The tooth exhibited a closed apex and an increased thickening of the canals. The pulp was necrotic but apically there was a periapical periodontitis meaning that HERS could still be viable and capable of physiological root formation once bacterial control was established even. The last one is from Peng *et al.*[52] (2017): bleeding was not achieved even though 2% lidocaine was used without vasoconstrictor. Glass ionomer cement was used as a capping material. After twelve months the tooth showed a normal physiological root development. The tooth diagnosis was irreversible pulpitis with periapical periodontitis. It is observed by the author that the tooth was previously treated with partial pulpectomy, these factors should have favoured the survival of HERS which could explain the success of the procedure. Also, upon histologic analysis, pulp-like and dentin-like tissue were found apically. It was not specified if the tooth was instrumented. Thus, the growing of those physiological tissues could be explained by remanent of pulp tissue.

Primary and secondary objectives

Periapical healing and resolution of the symptoms which is essential were observed in 95.7%. Apexogenesis, that here encompass either an increase in root length, thickness and/or signs of apical closure, was observed in 80.2%. Increase in root length reach 60%, root thickness 64.8% and apical closure 61,9%. Those results are coherent with the procedure as the 3-dimensional scaffold inside the canal produce a progressive obliteration of the canal following formation of mineralized tissue, which means that the increase in root thickness is slightly higher than the increase in root length. The high percentage of apical closure could also be explained as the increase in thickness of the root canal could lead to a closure of the apex by mineralized tissue without necessarily involving the increase in length.

Tertiary objective

Pulp vitality was found in 7.2% of the cases. This result, which is the tertiary goal of regenerative endodontic, could be misleading: the tissue growing inside the canal could be composed of periodontal ligament, cementum and bone, thus be vascularized, and elicit a positive response upon vitality test[25] without being proper pulp tissue. Another factor that could alter the vitality testing is the capping layer of MTA of usually 3 to 4mm which could give a false negative response[25]. Thus, without a proper histological confirmation the positive response to vitality test might not be considered as a true regeneration[20].

Discoloration

Discoloration represents 29.5% of the cases. While it was mainly due to minocycline present in the TAP[20], the capping material, whether it is grey MTA or white MTA, could also cause discoloration[53]. In this review, 7 cases reported discoloration after using TAP[26,29,39,46-48] and 1 changed its protocols to TAPm after ten teeth exhibited discoloration[35]. Following this modification in the protocol, no more teeth experienced discoloration. Another case experienced discoloration after the use of calcium hydroxide for three weeks followed by the placement of an Augmentin paste for 5 weeks[50]. The author did not give explanation about the possible etiology, but Augmentin has been associated with discoloration[54] together with calcium hydroxide[5]. Three other cases experienced discoloration even though they didn't use TAP with minocycline: Bakhtiar *et al.*[6] (2017), where the teeth suffered a minimal discoloration. Minocycline was replaced by cefaclor, discoloration was attributed to a possible interaction between Biodentine and PRF; Lin *et al.*[42] (2017) where minocycline was replaced by ciprofloxacin and the author used MTA as a capping material. No explanation about the discoloration were given by the author but it might be attributed to the use of MTA as discoloration was observed at the cervical part for all the affected teeth. And Kahler *et al.*[49] (2018), where amoxicillin was used instead of minocycline and white MTA as a capping material. The author reminds that from past studies, amoxicillin has been associated with discoloration too so it's suggested that it might be the cause. Other factors could be the use of white MTA as a capping material or the complete canal calcification that have affected the hydration of the tooth and the refraction of the light.

TABLE 2 · Pre-treatment

References	No. Of teeth	Tooth type	Avg. age	Sex	Aetiology	Pulp state	Periapical affectation	Instrumentation	Irrigation	Intracanal medication	Final irrigation	Evoked bleeding	Additional scaffold
Iwaya *et al.* (16)	1	PM	13	F	*	N	Y	Minimal	5% NaOCL 3% H2O2	DAP	*	*	*
Branchs e Trope (55)	1	PM	11	M	*	N	Y	N	5.25% NaOCL 0.12% CHX	TAP	*	*	*
Thibodeau e Trope (56)	1	I	9	M	T	N	Y	N	1.25% NaOCL	TAPm	*	Y	*
Cotti *et al.* (57)	1	I	9	F	T	N	Y	N	5% NaOCL 3% H2O2	Ca(OH)2	*	Y	*
Jung *et al.* (32)	4	4PM	10.75	4F	4*	4N	4Y	4*	2.5% NaOCL	3TAP 1Ca(OH)2	4*	4Y	4*
Thibodeau (58)	1	I	9	M	T	N	Y	N	1.25% NaOCL	TAPm	*	Y	*
Reynolds *et al.* (46)	2	2PM	11	F	2DE	2N	2Y	2*	6% NaOCL 2% CHX	2TAP	2*	2Y	2*
Ding *et al.* (59)	3	1I 2PM	8.5	2F 1M	1DE 2T	3*	3Y	3*	5.25% NaOCL	3TAP	3*	3Y	3*
Shin *et al.* (60)	1	PM	12	F	DI	N	Y	*	6% NaOCL 2% CHX	*	*	*	*
Petrino *et al.* (33)	2	2I 2PM	12	2M	2T 2*	4N	4Y	4*	5,25% NaOCL 0,12% CHX	4TAP	4*	2F 2Y	4*
Iwaya *et al.* (21)	1	I	8	M	T	N	Y	N	5% NaOCL 3% H2O2	Others	*	*	*
Jung *et al.* (34)	2	2PM	11	2F	2DE	2N	2Y	2*	2.5% NaOCL	Ca(OH)2 TAP	2*	1Y 1*	2*
Nosrat *et al.* (61)	2	2M	8,5	1F 1M	2C	2N	2Y	2N	5.25% NaOCL	2TAP	2*	2Y	2*
Miller *et al.* (47)	1	I	9	M	T	IP	Y	Y	2% CHX 17% EDTA	TAP	EDTA 17%	Y	*
Dabbagh *et al.* (62)	12	13I 1PM 2M	9.4	12*	12T 2C 1DE 1DI	16N	9Y 7*	16*	5% NaOCL	2TAP 14TAPm	16*	16Y	16*

References	No. Of teeth	Tooth type	Avg. age	Sex	Aetiology	Pulp state	Periapical affectation	Instrumentation	Irrigation	Intracanal medication	Final irrigation	Evoked bleeding	Additional scaffold
McTigue *et al.* (35)	29	26I 3PM	8,6	29*	25T 4DE	29N	29Y	29Y	3% NaOCL or 0,12% CHX	10TAP 19TAPc	29*	29Y	29*
Lenzi *et al.* (36)	1	2I	8	M	2T	2N	1Y 1*	2*	2.5% NaOCL	2TAP	2*	2Y	2*
Narayana *et al.* (37)	1	I	11	M	DI	N	Y	*	5.25% NaOCL	TAP	*	Y	*
Shimizu *et al.* (38)	1	I	9	M	T	IP	N	Minimal	5.25% NaOCL	Ca(OH)2	*	Y	*
Sönmez *et al.* (63)	3	3M	9	2F 1M	3C	3N	3Y	3*	5,25% NaOCL	3 TAP	3*	3Y	3*
Martin *et al.* (64)	1	M	9	M	C	N	Y	Y	5.25% NaOCL	TAP	*	Y	PRP
Soares *et al.* (65)	1	I	9	F	T	N	Y	Partial	2% CHX	Ca(OH)2 & 2% CHX	EDTA 17%	Y	*
Chen *et al.* (66)	1	PM	6	F	DE	N	Y	*	3% NaOCL CHX*	TAP	*	Y	*
Yang *et al.* (48)	1	I	11	M	DI	N	Y	N	5.25% NaOCL	TAP	*	F	*
Keswani *et al.* (51)	1	I	7	M	T	N	Y	N	5.25% NaOCL	TAP	*	N	*
Shimizu *et al.* (67)	1	I	9	M	T	N	Y	*	2.6 NaOCL	Ca(OH)2	*	Y	*
Nosrat *et al.* (50)	1	I	8	M	T	N	Y	Y	2,5% NaOCL	Ca(OH)2	*	Y	*
Bezgin *et al.* (44) metronidazole and cefaclor	2	2PM	12	2M	1C 1*	2N	2Y	2N	2.5% NaOCL 0.12% CHX	2TAPm	2EDTA 5%	2*	2PRP
Becerra *et al.* (68)	1	PM	11	F	1DE	N	T	N	5.25% NaOCL 2% CHX	TAP	*	Y	*
Lin *et al.* (39)	1	I	6	M	T	PIT	Y	N	5.25% NaOCL	TAP	*	Y	*
Nagy *et al.* (40)	20	20I	10.85	9M 11F	20*	20N	20*	20Minimal	2.6% NaOCL	20TAPd	20*	20Y	10 aHS + bFGF
Mccabe *et al.* (69)	1	I	7	F	T	N	Y	N	5% NaOCL	N	EDTA 17%	Y	*
Sachdeva *et al.* (70)	1	I	16	M	T	N	Y	N	5.25% NaOCL	TAP	*	*	PRP
Wang *et al.* (71)	1	2PM	39	F	2DE	2N	Y	*	2.5% NaOCL	2TAP	2*	2*	2PRP

References	No. Of teeth	Tooth type	Avg. age	Sex	Aetiology	Pulp state	Periapical affectation	Instrumentation	Irrigation	Intracanal medication	Final irrigation	Evoked bleeding	Additional scaffold
Ray *et al.* (72)	1	I	11	M	T	N	Y	*	0.5% NaOCL 17% EDTA	DAP	EDTA 17%	Y	PRF
Santiago *et al.* (26)	3	5I	8.6	3M	5T	5N	5Y	5*	5.25% NaOCL	5TAP	5*	5*	5*
Zhujiang *et al.* (73)	1	M	20	M	*	N	Y	Y	6% NaOCL 2% CHX	Ca(OH)2	EDTA 17%	Y	rhPDGF-BB
Li *et al.* (41)	20	20PM	10,6	11F 9M	20DE	20N	20Y	20N	2,5% NaOCL	20 Ca(OH)2	20*	20Y	20*
Timmerman *et al.* (7)	1	I	16	M	DI	PIT	Y	Minimal	1% NaOCL	Ca(OH)2	EDTA 15%	Y	*
Asgary *et al.* (54)	1	I	12	F	T	N	Y	Y	5.25% NaOCL	TAP	*	Y	*
Bakhtiar *et al.* (6)	4	5I	11	3F 1M	3PIT 1DI 1T	5N	4Y 1N	5Minimal	1.5% NaOCL	5TAPm	5 EDTA 17%	Y	PRF
Peng *et al.* (52)	1	PM	11	F	DE	IP	Y	*	5.25% NaOCL	TAP	*	F	*
Lin *et al.* (42)	69	21I 48PM	69*	69*	21T 48DE	69N	69Y	69*	1,25% NaOCL 17% EDTA	69TAPc	69 EDTA 17%	69Y	69N
Song *et al.* (43)29	29	7I 20PM 2M	12,4	17M 12F	14DE 2C 6T 7*	22N 5PIT 2IP	29Y	29*	2,5% NaOCL	9TAP 13 Ca(OH)2 4DAP 2 Ca(OH)2 + TAP 1 Ca(OH)2 + DAP	29*	23Y 6N	29*
Kahler *et al.* (49)	2	2PM	11	F	2*	2N	2Y	2*	1% NaOCL	2TAPa	2*	2Y	2*
Elsheshtawy *et al.* (29)	26	31I	12.65	15M 11F	30T 1DI	31N	18Y 13*	31Minimal	5.25% NaOCL	31TAP	31 EDTA 17%	17Y 14*	14 PRP

No, number; Avg, average; *, not specified in the article; I, incisor; C, canine; PM, premolar; M, molar; F, female; M, male; N, necrosis; IP, irreversible pulpitis; PIT, previously initiated therapy; Y, yes; N, no; F, failure; NaOCL, sodium hypochlorite; CHX, chlorhexidine; H2O2 , hydrogen peroxide: EDTA, ethylenediaminetetraacetic acid; DAP, double antibiotic paste – metronidazole, ciprofloxacin; TAP, triple antibiotic paste – metronidazole, ciprofloxacin, minocycline ; TAPm, triple antibiotic paste modified – metronidazole, ciprofloxacin, cefaclor; TAPd, triple antibiotic paste with doxycycline - metronidazole, ciprofloxacin, doxycycline; TAPa, triple antibiotic paste with amoxicillin – metronidazole, ciprofloxacin, amoxicillin; TAPc, triple antibiotic paste with clindamycin – metronidazole, ciprofloxacin, clindamycin; PRP, platelet rich plasma; PRF, platelet rich fibrin; aHS, artificial hydrogel scaffold; bFGF, basic fibroblast growth factor; rhPDGF-BB, recombinant human platelet derived growth factors.

TABLE 3 · Post-treatment

References	Average follow-up (in months)	Resolution of symptoms and periapical healing	Apexogenesis	Root length increase	Root width increase	Apical closure	Pulp vitality	Discoloration	Results
Iwaya *et al.* (16)	30	Y	Y	*	Y	Y	Y	*	S
Branchs e Trope (55)	24	Y	Y	Y	Y	Y	Y	*	S
Thibodeau e Trope (56)	12.5	Y	Y	Y	Y	Y	N	*	S
Cotti *et al.* (57)	30	Y	Y	Y	Y	Y	N	*	S
Jung *et al.* (32)	19.25	4Y	3Y 1N	1Y 1N 2*	3Y 1N	3Y 1N	4*	4*	3S 1F
Thibodeau (58)	41	Y	Y	Y	Y	Y	N	*	S
Reynolds *et al.* (46)	18	2Y	2Y	2Y	2Y	2Y	2Y	1Y 1N	2S
Ding *et al.* (59)	17	3Y	3Y	3Y	3Y	3Y	3Y	3*	3S
Shin *et al.* (60)	19	Y	Y	*	Y	Y	N	*	S
Petrino *et al.* (33)	12	4Y	3Y 1N	2Y 2N	3Y 1N	4N	2Y 2N	4*	3S 1F
Iwaya *et al.* (21)	156	Y	Y	*	Y	Y	Y	N	S
Jung *et al.* (34)	33.5	2Y	2*	1N 1*	1N 1*	2*	2*	2*	2F
Nosrat *et al.* (61)	16.5	2Y	2Y	1Y 1N	2Y	1Y 1N	2N	2*	2S
Miller *et al.* (47)	18	Y	Y	Y	Y	Y	Y	Y	S
Dabbagh *et al.* (62)	24	16Y	16Y	16Y	16Y	16Y	16*	2Y 14*	16S
McTigue *et al.* (35)	28,2	28Y 1N	22Y 7N	19Y 10N	20Y 9N	21Y 8N	29*	13Y 16N	22S 7F
Lenzi *et al.* (36)	21	2Y	1Y 1N	1Y 1*	1Y 1*	1Y 1*	2*	2*	1S 1F
Narayana *et al.* (37)	12	Y	*	N	N	*	*	*	F
Shimizu *et al.* (38)	1,5	*	*	*	*	*	*	*	F
Sönmez *et al.* (63)	24	3Y	3Y	3*	3Y	3Y	3*	3*	3S
Martin *et al.* (64)	25	Y	Y	*	Y	*	*	*	S
Soares *et al.* (65)	24	Y	Y	Y	Y	Y	N	*	S
Chen *et al.* (66)	12	Y	Y	Y	Y	Y	*	*	S

References	Average follow-up (in months)	Resolution of symptoms and periapical healing	Apexogenesis	Root length increase	Root width increase	Apical closure	Pulp vitality	Discoloration	Results
Yang *et al.* (48)	24	Y	Y	*	Y	Y	N	Y	S
Keswani *et al.* (51)	15	Y	Y	Y	Y	Y	Y	*	S
Shimizu *et al.* (67)	26	Y	Y	Y	Y	Y	*	*	S
Nosrat *et al.* (50)	31	Y	Y	N	Y	Y	N	Y	S
Bezgin *et al.* (44)	12	2Y	2Y	2*	2*	2Y	2N	2*	2S
Becerra *et al.* (68)	24	Y	Y	N	Y	Y	*	*	S
Lin *et al.* (39)	16	N	*	*	*	*	*	Y	F
Nagy *et al.* (40)	18	17Y 3*	17Y 3*	17Y 3*	17Y 3*	17Y 3*	20*	20*	17S 3F
Mccabe *et al.* (69)	18	Y	Y	Y	Y	Y	*	*	S
Sachdeva *et al.* (70)	36	Y	Y	Y	Y	Y	N	*	S
Wang *et al.* (71)	30	2Y	2N	2N	2N	2N	2N	2*	2F
Ray *et al.* (72)	36	Y	Y	Y	N	N	N	Y	S
Santiago *et al.* (26)	63	1Y 4N	1Y 4N	1Y 4N	1Y 4N	1Y 4N	5*	1Y 4N	5F
Zhujiang *et al.* (73)	15	Y	Y	*	Y	Y	Y	*	S
Li *et al.* (41)	12	18Y 2N	11Y 9N	11Y 9N	20*	8Y 12N	5Y 15N	20*	11S 9F
Timmerman *et al.* (7)	36	Y	Y	*	Y	Y	N	*	S
Asgary *et al.* (54)	36	Y	Y	Y	Y	Y	*	*	S
Bakhtiar *et al.* (6)	18	5Y	5Y	5*	5Y	4Y 1*	5*	3Y 2*	5S
Peng *et al.* (52)	12	Y	Y	Y	Y	Y	*	*	S
Lin *et al.* (42)	12	69Y	57Y 12*	56Y 13*	57Y 12*	46Y 23*	69*	30Y 39*	57Y 12F
Song *et al.* (43)29	24,9	29Y	23Y 6N	23Y 6N	23Y 6N	23Y 6N	29*	29*	23S 6F
Kahler *et al.* (49)	96	2Y	2Y	2N	2Y	2*	2Y	2Y	2S
Elsheshtawy *et al.* (29)	12	31Y	27Y 4N	31*	31*	31*	31*	25Y 6*	27S 4F

TABLE 4 · Demographic and study data

Variable	%
Sex	
Male	30.38%
Female	27.31%
Not specified	42.31%
Tooth type	
Incisor	54.32%
Premolar	41.73%
Molar	3.96%
Aetiology	
Trauma	43.53%
Dens invaginatus	2.52%
Dens evaginatus	34.89%
Caries	3.96%
Others	1.08%
Not specified	14.03%
Pulp state	
Necrosis	94.6%
Irreversible pulpitis	1.8%
Others	2.52%
Not specified	1.08%
Periapical state	
Apical periodontitis	46.76%
Apical abscess	18.71%
Normal periapical tissue	0.72%
Not precise enough	26.26%
Not specified	7.55%
Instrumentation	
No	12.59%
Yes	12.23%
Partial	0.36%
Minimal	21.22%
Not specified	53.6%
Irrigant	
NaOCL	65.22%
NaOCL with other irrigants	30.43%
Other combinations	4.35%
Intracanal medication	
TAP	32.73%
TAPm	8.27%
DAP	2.16%
Ca(OH)2	14.75%
Other combinations	41.37%
None	0.36%
Not specified	0.36%

TABLE 5 · Follow-up variables

Variable	Yes (%)	No (%)	Not specified (%)
Periapical healing	95.68%	2.88%	1.44%
Apexogenesis	80.22%	12.59%	7.19%
Increase root length	60.07%	14.75%	25.18%
Increase root thickness	64.75%	9.35%	25.9%
Apical closure	61.87%	14.03%	24.1%
Pulp vitality	7.19%	11.87%	80.94%
Discoloration	29.5%	7.91%	62.59%

Conclusion

Regarding the present study, regenerative endodontic showed signs of apexogenesis in the majority of cases, and a pronounced increase in root thickness, root length together with the closure of the apex in a great number of teeth. Those results demonstrate the great variability in procedures and outcomes regardless of the applied protocols. Due to a high number of protocols and their heterogenicity, the superiority of one over the others cannot be established. More randomized clinical trials are needed to determine the most effective protocol. This study encloses a majority of case reports and thus they can be subject to publication bias, which is an issue. Finally, all the studies were not analysed with the same rigour by their author due to the lack of standardized analysing methods.

A guideline which lists the different information that the dentist must follow should be established for a better understanding and analysis of each case. The precise role of stem cells in regenerative endodontic should be studied more in depth to guide the clinicians to the need of bacterial-free microenvironment while preserving stem cells.

Highlights

- Regenerative endodontic is one of the most promising procedures among the field of endodontic treatments.
- Regenerative endodontic have the potential to induce further root maturation in teeth with arrested root development.
- Discoloration together with intracanal calcifications are possible challenging adverse effects for the dentist that must be considered before electing the procedure.
- Numerous protocols are used among clinical cases with varying results which impede to establish the superiority of one over the others.

References

1 - Singh RK, Kumar SR, Kumar SV, Khanna R, Singh BP, Jindal G, *et al.* Cochrane Database of Systematic Reviews Interventions for managing immature permanent teeth with necrotic pulps (Protocol). Cochrane Database Syst Rev. 2017; 6(6): CD012709.

2 - Diogenes A, Ruparel NB, Shiloah Y, Hargreaves KM. Regenerative endodontics A way forward. J Am Dent Assoc. 2016; 147(5): 372–80.

3 - Vishwakarma A, Sharpe P, Shi S, Ramalingam M. Stem Cell Biology and Tissue Engineering in Dental Sciences. Elsevier: Amsterdam; 2014; p. 1-900.

4 - Huang GT, Sonoyama W, Liu Y, Liu H, Wang S, Shi S. The Hidden Treasure in Apical Papilla: The Potential Role in Pulp/ Dentin Regeneration and BioRoot Engineering. J Endod. 2008; 34(6): 645–651.

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

6 - Bakhtiar H, Esmaeili S, Fakhr Tabatabayi S, Ellini MR, Nekoofar MH, Dummer PMH. Second-generation platelet concentrate (platelet-rich fibrin) as a scaffold in regenerative endodontics: a case series. J Endod. 2017; 43(3): 401–8.

7 - Timmerman A, Parashos P. Delayed Root Development by Displaced Mineral Trioxide Aggregate after Regenerative Endodontics: A Case Report. J Endod. 2017; 43(2): 252–6.

8 - Chen YP, Jovani-Sancho MM, Sheth CC. Is revascularization of immature permanent teeth an effective and reproducible technique? Dent Traumatol. 2015; 31(6): 429–36.

9 - Lin LM, Kahler B. A review of regenerative endodontics: current protocols and future directions. J Istanbul Univ Fac Dent. 2017; 51:41–51.

10 - Diogenes AR, Ruparel NB, Teixeira FB, Hargreaves KM. Translational science in disinfection for regenerative endodontics. J Endod. 2014; 40(4 Suppl.):S52–7.

11 - Chubb DWR. A review of the prognostic value of irrigation on root canal treatment success. Aust Endod J. 2019; 45(1):5-11.

12 - Zehnder M. Root Canal Irrigants. J Endod. 2006; 32(5): 389–98.

13 - Sim TPC, Knowles JC, Ng YL, Shelton J, Gulabivala K. Effect of sodium hypochlorite on mechanical properties of dentine and tooth surface strain. Int Endod J. 2001; 34(2): 120–32.

14 - Martin DE, Almeida JFA, Henry MA, Khaing ZZ, Schmidt CE, Teixeira FB, *et al.* Concentration-dependent effect of sodium hypochlorite on stem cells of apical papilla survival and differentiation. J Endod. 2014; 40(1):51–5.

15 - Demarco FF, Casagrande L, Zhang Z, Dong Z, Tarquinio SB, Zeitlin BD, *et al.* Effects of morphogen and scaffold porogen on the differentiation of dental pulp stem cells. J Endod. 2010; 36:1805-11.

16 - Iwaya SI, Ikawa M, Kubota M. Revascularization of an immature permanent tooth with apical periodontitis and sinus tract. Dent Traumatol. 2001; 17(4): 185-7.

17 - Kontakiotis EG, Filippatos CG, Tzanetakis GN, Agrafioti A. Regenerative endodontic therapy: a data analysis of clinical protocols. J Endod. 2015; 41(2):146–54.

18 - Nosrat A, Homayounfar N, Oloomi K. Drawbacks and unfavorable outcomes of regenerative endodontic treatments of necrotic immature teeth: a literature review and report of a case. J Endod. 2012; 38(10): 1428-34.

19 - Ruparel NB, Teixeira FB, Ferraz CCR, Diogenes A. Direct effect of intracanal medicaments on survival of stem cells of the apical papilla. J Endod. 2012; 38(10): 1372–5.

20 - Galler KM. Clinical procedures for revitalization: current knowledge and considerations. Int Endod J. 2016; 49(10): 926-36.

21 - Iwaya SI, Ikawa M, Kubota M. Revascularization of an immature permanent tooth with periradicular abscess after luxation. Dent Traumatol. 2011; 27(1): 55-8.

22 - Graham L, Cooper PR, Cassidy N, Nor JE, Sloan AJ, Smith AJ. The effect of calcium hydroxide on solubilisation of bio-active dentine matrix components. Biomaterials. 2006; 27(14): 2865-73.

23 - Yassen GH, Vail MM, Chu TG, Platt JA. The effect of medicaments used in endodontic regeneration on root fracture and microhardness of radicular dentine. Int Endod J. 2013; 46(7): 688-95.

24 - Kim SG, Zhou J, Solomon C, Zheng Y, Suzuki T, Chen M, *et al.* Effects of Growth Factors on Dental Stem/Progenitor Cells. Dent Clin North Am. 2012; 56(3):563-75.

25 - Bezgin T, Sönmez H. Review of current concepts of revascularization/revitalization. Dent Traumatol. 2015; 31(4): 267-73.

26 - Santiago CN, Pinto SS, Sassone LM, Hirata R, Fidel SR. Revascularization technique for the treatment of external inflammatory root resorption: a report of 3 cases. J Endod. 2015; 41(9): 1560-4.

27 - Ulusoy AT, Turedi I, Cimen M, Cehreli ZC. Evaluation of blood clot, platelet-rich plasma, platelet-rich fibrin, and platelet pellet as scaffolds in regenerative endodontic treatment: a prospective randomized trial. J Endod. 2019; 45(5): 560–6.

28 - American Association of Endodontists. AAE Clinical Considerations for a Regenerative Procedure. [online]. Available from: https://www.aae.org/specialty/wp-content/uploads/sites/2/2018/06/ConsiderationsForRegEndo_AsOfApril2018.pdf.

29 - ElSheshtawy AS, Nazzal H, El Shahawy OI, El Baz AA, Ismail SM, Kang J, *et al.* The effect of platelet-rich plasma as a scaffold in regeneration/revitalization endodontics of immature permanent teeth assessed using 2-dimensional radiographs and cone beam computed tomography: a randomized controlled trial. Int Endod J. 2020; 53(7): 905-21.

30 - Garcia-Godoy F, Murray PE. Recommendations for using regenerative endodontic procedures in permanent immature traumatized teeth. Dent Traumatol. 2012; 28(1): 33-41.

31 - Estefan BS, El Batouty KM, Nagy MM, Diogenes A. Influence of Age and Apical Diameter on the Success of Endodontic Regeneration Procedures. J Endod. 2016; 42(11): 1620-5.

32 - Jung IY, Lee SJ, Hargreaves KM. Biologically based treatment of immature permanent teeth with pulpal necrosis: a case series. J Endod. 2008;34(7): 876-87.

33 - Petrino JA, Boda KK, Shambarger S, Bowles WR, McClanahan SB. Challenges in regenerative endodontics: a case series. J Endod. 2010; 36(3): 536-41.

34 - Jung IY, Kim ES, Lee CY, Lee SJ. Continued development of the root separated from the main root. J Endod. 2011; 37(5): 711-4.

35 - McTigue DJ, Subramanian K, Kumar A. Management of immature permanent teeth with pulpal necrosis: a case series. Pediatr Dent. 2013;35(1): 55-60.

36 - Lenzi R, Trope M. Revitalization procedures in two traumatized incisors with different biological outcomes. J Endod. 2012; 38(3): 411-4.

37 - Narayana P, Hartwell GR, Wallace R, Nair UP. Endodontic clinical management of a dens invaginatus case by using a unique treatment approach: A case report. J Endod. 2012; 38(8): 1145-8.

38 - Shimizu E, Jong G, Partridge N, Rosenberg PA, Lin LM. Histologic observation of a human immature permanent tooth with irreversible pulpitis after revascularization/regeneration procedure. J Endod. 2012; 38(9): 1293-7.

39 - Lin LM, Shimizu E, Gibbs JL, Loghin S, Ricucci D. Histologic and histobacteriologic observations of failed revascularization/revitalization therapy: a case report. J Endod. 2014; 40(2): 291-5.

40 - Nagy MM, Tawfik HE, Hashem AAR, Abu-Seida AM. Regenerative potential of immature permanent teeth with necrotic pulps after different regenerative protocols. J Endod. 2014; 40(2): 192-8.

41 - Li L, Pan Y, Mei L, Li J. Clinical and Radiographic Outcomes in Immature Permanent Necrotic Evaginated Teeth Treated with Regenerative Endodontic Procedures. J Endod. 2017;43(2): 246–51.

42 - Lin J, Zeng Q, Wei X, Zhao W, Cui M, Gu J, *et al.* Regenerative endodontics versus apexification in immature permanent teeth with apical periodontitis: a prospective randomized controlled study. J Endod. 2017; 43(11): 1821-7.

43 - Song M, Cao Y, Shin SJ, Shon WJ, Chugal N, Kim RH, *et al.* Revascularization-associated Intracanal Calcification: Assessment of Prevalence and Contributing Factors. J Endod. 2017; 43(12): 2025-33.

44 - Bezgin T, Yilmaz AD, Çelik BN, Sönmez H. Concentrated platelet-rich plasma used in root canal revascularization: 2 case reports. Int Endod J. 2014; 47(1): 41-9.

45 - Nagata JY, Gomes BP, Rocha Lima TF, Murakami LS, Faria DE, Campos GR, Souza-Filho FJ, Soares AJ. Traumatized immature teeth treated with 2 protocols of pulp revascularization. J Endod. 2014; 40(5): 606–12.

46 - Reynolds K, Johnson JD, Cohenca N. Pulp revascularization of necrotic bilateral bicuspids using a modified novel technique to eliminate potential coronal discolouration: A case report. Int Endod J. 2009; 42(1):84–92.

47 - Miller EK, Vann WF. Emerging therapies for the management of traumatized immature permanent incisors. Pediatr Dent. 2010; 34(1):66–70.

48 - Yang J, Zhao Y, Qin M, Ge L. Pulp revascularization of immature dens invaginatus with periapical periodontitis. J Endod. 2013; 39(2): 288–92.

49 - Kahler B, Kahler SL, Lin LM. Revascularization-associated Intracanal Calcification: A Case Report with an 8-year Review. J Endod. 2018; 44(12): 1792–5.

50 - Nosrat A, Li KL, Vir K, Hicks ML, Fouad AF. Is pulp regeneration necessary for root maturation? J Endod. 2013; 39(10): 1291-5.

51 - Keswani D, Pandey RK. Revascularization of an immature tooth with a necrotic pulp using platelet-rich fibrin: a case report. Int Endod J. 2013; 46(11): 1096-104.

52 - Peng C, Zhao Y, Wang W, Yang Y, Qin M, Ge L. Histologic findings of a human immature revascularized/regenerated tooth with symptomatic irreversible pulpitis. J Endod. 2017; 43(6): 905-9.

53 - Kahler B, Mistry S, Moule A, Ringsmuth AK, Case P, Thomson A, *et al.* Revascularization outcomes: a prospective analysis of 16 consecutive cases. J Endod. 2014; 40(3): 333-8.

54 - Asgary S, Fazlyab M, Nosrat A. Regenerative endodontic treatment versus apical plug in immature teeth: Three-year follow-up. J Clin Pediatr Dent. 2016; 40(5): 356-60.

55 - Banchs F, Trope M. Revascularization of immature permanent teeth with apical periodontitis: new treatment protocol? J Endod. 2004; 30(4): 196-200.

56 - Thibodeau B, Trope M. Pulp Revascularization of a necrotic Infected immature permanent tooth: Case report ande review. Pediatr Dent. 2007; 29(1): 47-50.

57 - Cotti E, Mereu M, Lusso D. Regenerative treatment of an immature, traumatized tooth with apical periodontitis: report of a case. J Endod. 2008; 34(5): 611-6.

58 - Thibodeau B. Case report: pulp revascularization of a necrotic, infected, immature, permanent tooth. Pediatr Dent. 2009; 31(2): 145-8.

59 - Ding RY, Cheung GS pan, Chen J, Yin XZ, Wang QQ, Zhang CF. Pulp revascularization of immature teeth with apical periodontitis: a clinical study. J Endod. 2009; 35(5): 745-9.

60 - Shin SY, Albert JS, Mortman RE. One step pulp revascularization treatment of an immature permanent tooth with chronic apical abscess: a case report. Int Endod J. 2009; 42(12): 1118-26.

61 - Nosrat A, Seifi A, Asgary S. Regenerative endodontic treatment (revascularization) for necrotic immature permanent molars: a review and report of two cases with a new biomaterial. J Endod. 2011; 37(4): 562-7.

62 - Dabbagh B, Alvaro E, Vu DD, Rizkallah J, Schwartz S. Clinical complications in the revascularization of immature necrotic permanent teeth. Pediatr Dent. 2012; 34(5): 414-7.

63 - Sönmez IS, Akbay OA, Erkmen AM. Revascularization/regeneration performed in immature molars: case reports. J Clin Pediatr Dent. 2013; 37(3): 231-4.

64 - Martin G, Ricucci D, Gibbs JL, Lin LM. Histological findings of revascularized/revitalized immature permanent molar with apical periodontitis using platelet-rich plasma. J Endod. 2013; 39(1): 138-44.

65 - Soares ADJ, Lins FF, Nagata JY, Gomes BPFDA, Zaia AA, Ferraz CCR, *et al.* Pulp revascularization after root canal decontamination with calcium hydroxide and 2% chlorhexidine gel. J Endod. 2013; 39(3): 417-20.

66 - Chen X, Bao ZF, Liu Y, Liu M, Jin XQ, Xu XB. Regenerative endodontic treatment of an immature permanent tooth at an early stage of root development: A case report. J Endod. 2013; 39(5): 719-22.

67 - Shimizu E, Ricucci D, Albert J, Alobaid AS, Gibbs JL, Huang GTJ, *et al.* Clinical, radiographic, and histological observation of a human immature permanent tooth with chronic apical abscess after revitalization treatment. J Endod. 2013; 39(8): 1078-83.

68 - Becerra P, Ricucci D, Loghin S, Gibbs JL, Lin LM. Histologic study of a human immature permanent premolar with chronic apical abscess after revascularization/revitalization. J Endod. 2014; 40(1):133-9.

69 - Mccabe P. Revascularization of an immature tooth with apical periodontitis using a single visit protocol: a case report. Int Endod J. 2015; 48(5): 484–97.

70 - Sachdeva GS, Sachdeva LT, Goel M, Bala S. Regenerative endodontic treatment of an immature tooth with a necrotic pulp and apical periodontitis using platelet-rich plasma (PRP) and mineral trioxide aggregate (MTA): a case report. Int Endod J. 2015; 48(9): 902-10.

71 - Wang Y, Zhu X, Zhang C. Pulp revascularization on permanent teeth with open apices in a middle-aged patient. J Endod. 2015; 41(9): 1571-5.

72 - Ray HL, Marcelino J, Braga R, Horwat R, Lisien M, Khaliq S. Long-term follow up of revascularization using platelet-rich fibrin. Dent Traumatol. 2016; 32(1): 80-4.

73 - Zhujiang A, Kim SG. Regenerative endodontic treatment of an immature necrotic molar with arrested root development by using recombinant human platelet-derived growth factor: a case report. J Endod. 2016; 42(1): 72–5.

Evolução da endodontia regenerativa em dentes permanentes jovens: uma revisão da literatura

Resumo

Objetivo: comparar diferentes protocolos utilizados em endodontia regenerativa. Metodologia: a busca foi feita em artigos publicados entre 2001 e 2021 no PubMed usando as palavras-chave: "pulp revascularization" ou "pulp revitalization" ou "apexification". Os critérios de inclusão foram estudos em humanos, pacientes com dentes permanentes imaturos com necrose ou pulpite irreversível, acompanhamento de pelo menos 12 meses, protocolos claramente descritos e em inglês. Resultados: A busca resultou em 944 artigos. Após aplicação dos critérios de inclusão e exclusão, 35 foram selecionados. 11 artigos foram adicionados após uma revisão bibliográfica dos artigos selecionados e um total de 46 artigos foram incluídos. A taxa de sucesso para casos de endodontia regenerativa foi de 80% observada em 278 dentes. Durante a irrigação, o NaOCL foi utilizado na maioria dos casos, com concentrações variando de 0,5% a 6% e a TAP foi a medicação intracanal mais utilizada (32,7%). O reparo periapical e remissão dos sintomas foram observados em 95,7% dos casos, enquanto a apexigênese foi observada em 80,2%. Em 60% dos casos houve aumento no comprimento da raiz, em 64,8% da espessura da raiz e fechamento apical em 61,9%. Vitalidade pulpar foi encontrada em 7,2% dos casos e descoloração em 29,5%. Conclusão: A endodontia regenerativa mostrou sinais de apexigênese na maioria dos casos, e um aumento pronunciado na espessura da raiz, comprimento da raiz e fechamento do ápice. Devido ao grande número de protocolos e sua heterogeneidade, não se pode estabelecer a superioridade de um sobre os outros. Mais ensaios clínicos randomizados são necessários para determinar o protocolo mais eficaz.

Palavras-chave: Endodontia Regenerativa; Apexificação; Cavidade Pulpar; Desinfecção.

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