Abstract

Objectives: This study aims to review all the available strategies and treatment protocols used for regeneration of necrotic immature roots in the published reported clinical cases. Methods/ Statistical Analysis: The following electronic bibliographic databases; Pubmed, Scirus, Cochrane Library and Summon were searched, in addition to “hand searching” to identify other potentially relevant articles. A structured electronic search was carried out including; only terms related to the intervention. Relevant papers published in English were identified after reviewing their titles, abstracts then full reading of the papers. Findings: 32 papers were included according to the inclusion criteria. They were directly relevant to the subject; all of them are clinical studies following regenerative endodontic therapy to treat the necrotic immature teeth. Application/ Improvement: Different methodological steps are followed to regenerate the necrotic immature roots. Reviewing these variability and the different strategies followed was an important issue to reach evidence based guidelines and showed the clinical significance for application in the daily clinical practice.

Keywords: Evidence-Based Medicine, Maturogenesis, Pulp Revitalization, Systematic Review, Young Permanent Teeth

1. Research Question

What are the strategies used for regeneration of young permanent teeth with necrotic pulp?

P: Necrotic young permanent teeth
I: Regenerative endodontic treatment
O: Primary outcome is root development and resolution of signs and symptoms

2. Introduction

One of the worst scenarios in children dentition is a necrotic pulp of a young permanent tooth. Losing the pulp before completion of root formation will lead to the following cons; short root, wide apex and thin root walls. An endodontic root canal treatment is required to maintain this tooth; first of all controlling the infection, second; creating an apical tight seal. These aims offered by calcium hydroxide apexification or by creating an instant apical barrier using Mineral Trioxide Aggregate (MTA). But a problem still exists which is a short root with thin root walls; liable to future fractures. A paradigm shift for treatment of such cases emerges by the development of tissue engineering technologies using stem cells; regeneration or commonly called revitalization, revascularization or maturogenesis. Regeneration changes the necrotic root into a vital root able to complete its development. Regeneration not only closes the wide apex as that by the apexification technique but also increases root wall thickness and root length. A stronger root with an adequate crown-root ratio is obtained; which is more resistant to future fracture.

Saving immature teeth is a crucial step in preserving the alveolar bone of a growing child and keeping his smile. Revascularization of non-vital teeth allows repair and regeneration of tissues. The rationale of revascularization is that pulp vitality can be reestablished if a sterile tissue matrix is provided in which new cells can grow. Components needed for tissue engineering; stem cells,
signaling molecules and scaffold. The first case reports for revitalizing a necrotic pulp were published in 2001 by and then followed by many case reports, case series, animal studies, narrative reviews and only one systematic review. There isn’t any randomized controlled trial published yet.

3. Objectives

This systematic review aims to review all the available strategies and treatment protocols used for regeneration of necrotic immature roots in the published reported clinical cases.

4. Search Strategy

We searched the following electronic bibliographic databases: Pubmed, Scirus, The Cochrane Library and Summon. In addition to hand searching of the identified journals' indexes, special issues, bibliographies and reference lists of identified articles were scanned to identify other potentially relevant articles. Structured electronic search was carried out including only terms relating to the intervention. The search terms are revascularization pulp - maturogenesis - pulp revitalization - regenerative endodontic treatment - regenerative endodontic procedures - regenerative endodontics - revascularization in regenerative endodontics - regenerative endodontic - challenges in regenerative endodontics - regenerative root canal treatment - regenerative root canal therapy - pulp revascularization. The search was last updated on 28 January 2015. Papers in the English language are the only ones included.

URL to search strategy in Pubmed: http://www.ncbi.nlm.nih.gov/pubmed/advanced, (revascularization pulp OR maturogenesis) OR pulp revitalization) OR regenerative endodontic treatment) OR regenerative endodontic procedures) OR regenerative endodontics) OR revascularization in regenerative endodontics) OR regenerative endodontic) OR challenges in regenerative endodontics) OR regenerative root canal treatment) OR regenerative root canal therapy) OR pulp revascularization

Criteria for including and excluding studies in the review: Table 1 is showing the inclusion and exclusion criteria for considering studies in the review based on the PIO structure

| Inclusion criteria | Exclusion Criteria |
|--------------------|--------------------|
| Population         | Outcome            |
| Necrotic immature  | Clinical evaluation for signs and symptoms |
| teeth              | Radiographic evaluation for periapical tissues and root development |
| Teeth with fractured roots | Histological evaluation |

| Type of study | Inclusion criteria | Exclusion Criteria |
|---------------|--------------------|--------------------|
| Clinical trials | Animal studies |
| Case reports   | In-vitro studies |
| Case series    | Retrospective studies |
| In-vivo studies | Histological studies |

Results: Figure 1 is showing the results of the search strategy in pubmed, summon, cochrane and scirus jour-}

5. Methods of Review

5.1 Data extraction Strategy

Titles and abstracts of studies retrieved using the search strategy and those from additional sources were screened independently by the two review authors to identify studies that potentially meet the inclusion criteria outlined above. The full text of these potentially included studies were retrieved and independently assessed for eligibility by the two review team members.

Table 1. Inclusion and exclusion criteria for considering studies in the review based on the PIO structure

Figure 1. Flow diagram for the search results
A standardized, pre-piloted form used to extract data from the included studies for assessment of study quality and evidence synthesis. Extracted information include: demographic data, methodological steps and results as shown in Table 2.

6. Critical Appraisal for the Included Studies

A standardized assessment of methodological quality of each paper is carried out to improve inter and intra rater reliability by applying the critical appraisal approach used by the Oxford Centre for Evidence Medicine. A 10-question checklist was completed for each study http://www.cebma.org/wp-content/uploads/Critical-Appraisal-Questions-for-a-Case-Study.pdf. Any disagreement was solved by discussion.

**Demographic Data:** Table 3 is showing the demographic data of the included studies in terms of; treated teeth, patient age and gender.

| Ref. | Treated teeth | Patient age | Gender | Dental History of Present Illness |
|------|---------------|-------------|--------|----------------------------------|
| 1    | premolar      | 13yrs       | Female | The tooth was free of caries, but with fracture of the central cusp |
| 27   | premolar      | 11yrs       | Male   | The tooth appeared intact, without caries. The presence of occlusal tubercles on the other mandibular premolars suggested that one may have been present on this tooth, which was fractured during function, resulting in a microexposure and necrosis of the pulp. The tooth had an open apex associated with a large radiolucency and a lingual sinus tract |
| 12, 13 | incisor     | 9yrs        | Male   | Tooth with complicated crown fracture and acute apical abscess with history of dental trauma 2 years ago and previously treated with Cvekpulpotomy approximately 48 hours after the accident without rubber dam isolation |
| 24   | 9 premolars   | 9-14yrs     | 4Males 4Females | Teeth with pulp necrosis and apical periodontitis |
| 31   | 14 teeth      | 9-18yrs     | 9 Males 5 Female | Teeth presenting with or without signs and/or symptoms of periapical pathology, fractured or discolored anterior teeth and were associated with symptoms of acute or chronic apical infection, i.e, diffuse facial swelling or intraoral sinuses. |
| 35   | 5 incisors 7 premolars | 8-11yrs | 5 Males 7 Female | Teeth with or without signs and/or symptoms of periapical pathology The patients presented with either a fractured dens invaginatus or traumatized teeth |
| 15   | 2 premolars   | 11yrs       | Female | Teeth with or without signs and/or symptoms of periapical pathology Replanted upper centrals showing pulpal necrosis and asymptomatic apical periodontitis |
| 9    | 1 premolar    | 12yrs       | Female | Asymptomatic tooth exhibited occlusal caries with an associated draining sinus tract |
| 23   | 4 incisors 2 premolar | 6-11yrs | 2 Males 1 Female | Broken upper centrals due to a fall nearly 6 years ago Tooth with asymptomatic apical periodontitis and tooth with chronic apical abscess Replanted upper centrals showing pulpal necrosis and asymptomatic apical periodontitis |
| 18   | 1 premolar    | 12yrs       | Female | Tooth suffered from acute apical periodontitis and mobility after periodontal treatment, after 2 weeks from splinting; gingival swelling and abscess formation |
| 19   | 1 incisor     | 7yrs        | Male   | Tooth suffered from acute apical periodontitis and mobility after periodontal treatment, after 2 weeks from splinting; gingival swelling and abscess formation |
| Patient | Teeth | Age/Group | Sex | Diagnosis/Details |
|---------|-------|-----------|-----|-------------------|
| 17      | 2 molars | 8-9yrs | Male | Broken tooth-colored restoration and extensive caries, with localized swelling in the buccal mucosa. Extraoral and intraoral swelling related to molar with extensive caries. |
| 16      | 6 molars | 8-11yrs | ___ | No signs of swelling or sinus tracts. 4 molars undergo previous instrumentation. |
| 6, 7    | 1 premolar | 11yrs | Male | Tooth was accidentally extracted and immediately replanted, had pulpal necrosis, symptomatic apical periodontitis and slightly discoloration. |
| 8       | 3 premolars | 12 yrs/10 yrs | 2 Males | Pulp necrosis with symptomatic apical periodontitis. Acute swelling and severe pain. Slight gingival swelling, sensitivity to percussion and negative response to electric pulp test. |
| 11      | 1 incisor | 8yrs | Male | Tooth presented with acute apical abscess with buccal swelling, history of trauma 6 months ago. |
| 42      | 18 teeth | 7-15.5 yrs | ___ | 14 were the result of trauma, 2 had been caused by caries, and 2 originated from dental anomalies (dens evaginatus and dens invaginatus). |
| 28      | 10 incisors/10 premolars | 8-13yrs | 4 Males/6 Female | Incisors had history of trauma and pulp infection. 3 premolars had caries with subsequent pulp involvement. 7 premolars with dens invaginatus and pulp involvement. 12 teeth had a draining sinus tract. |
| 21      | 2 incisors | 8yrs | Male | Medical history revealed the child was under cardiac observation after a surgical repair for an interventricular communication at age 5. But he was in a very good health, teeth show crown fracture after a fall at school 2.5 months ago. |
| 22      | 2 incisors | 8.5yrs | Male | The upper central incisors showed severe extrusive luxation, excessive mobility, a fractured labial cortical bone and slightly mobile palatal segment of the alveolar bone. |
| 29      | incisor | 24yrs | Female | History of trauma 15 years ago, patient suffer from pain, swelling and pus discharge in relation to upper front region, the central incisors had grade I mobility. |
| 10      | 1 incisor | 9yrs | Male | Broken tooth due to trauma along with discoloration. |
| 30      | 20 teeth | 15-28 yrs | 14 Males/6 Female | Patients having nonvital, immature anterior teeth with or without radiographic evidence of periapical lesions. |
| 26      | 2 incisors | 14yrs | Female | History of impact trauma to the anterior maxillary teeth 6 years ago, extensive caries and complicated crown fracture of tooth and uncomplicated fracture of the other tooth, the patient’s chief complaints were repeated swelling and pain in the anterior maxilla. |
| 32      | 1 incisor | 9yrs | Female | Enamel-dentin-pulp fracture in the tooth associated with intrusive luxation that was pulp capped and after 3 weeks negative responses to cold pulp test and EPT. |
| 34      | 1 incisor | 17yrs | Male | Carious tooth with history of trauma 8 years ago. |
| 14      | 1 incisor | 11yrs | Male | Patient with history of systemic compromise, tooth is cone shaped without caries, with type II invagination extending from the crown to the middle root and no history of dental trauma, Swelling of the palatal gingival. |
| 20      | 1 incisor | 7yrs | Male | History of trauma 5 months ago, painful tooth with Class IV fracture and acute apical periodontitis. |
| 33      | 1 incisor | 11yrs | Male | History of trauma one month ago, tooth with Ellis class III fracture and mobility. |
| 40      | 2 premolars | 12yrs | 2 Males | The tooth had deep carious lesion without any clinical signs or symptoms or radiographic evidence for periapical pathosis; was diagnosed with a necrotic pulp. Tooth with composite restoration showing pain, sinus tract, tenderness to percussion and palpation. |
Table 4. Methodological steps

| Ref. | Study Design | Irrigants Used | Instrumentation Or Additional steps | Antibiotic Paste | Controlling the infection inside the root canal lasts for | Scaffold Used | Coronal Seal |
|------|--------------|----------------|-------------------------------------|------------------|----------------------------------------------------------|-------------|-------------|
| 1    | Case report  | 5% NaOCl 3% H2O2 | the root canal was not mechanically cleaned to its full length | Metronidazole +Ciprofloxacin +Minocycline | Not mentioned | No induced bleeding | Calcium hydroxide paste +Glass Ionomer cement + Adhesive resin restoration |
| 27   | Case report  | 20 ml 5.25% NaOCl +10 ml of Peridex | No instrumentation | Metronidazole +Ciprofloxacin +Minocycline | 26days | Blood clot | MTA + Adhesive resin restoration |
| 12, 13 | Case report | 10 ml of 1.25% NaOCl | No Instrumentation | Metronidazole +Ciprofloxacin +Cefaclor | 11weeks | Blood clot | MTA + Adhesive resin restoration |
| 24   | Case series  | 5.25% NaOCl for 10 minutes 2.5% NaOCl was continued for 30 minutes | No instrumentation | Metronidazole +Ciprofloxacin +Minocycline then mixture of erythromycin +Ca(OH)2 Metronidazole +Ciprofloxacin +Minocycline Ca(OH)2 only | Minimum 1 week up to | Without inducing bleeding. There seemed to be some vital tissue remaining in the apical half of the canal because insertion of a K-file evoked a sensation of pain. Blood clot | Ca(OH)2 paste After calcific bridge formation; Obtura II and adhesive resin restoration MTA + Adhesive resin restoration MTA+IRM Collatape +MTA + Adhesive resin restoration |
| 31   | A pilot Clinical study | 3% H2O2 + 2.5% NaOCl | Minimal instrumentation | Formocresol was used as an inter appointment dressing | | Blood clot | Glass Ionomer |
| 35   | Clinical study | 20 ml of 5.25% NaOCl | Metronidazole +Ciprofloxacin +Minocycline | | the tooth was left open to drain for 3 days after which dressed with the antibiotic mixture for 1 week | Blood clot in 6 teeth out of 12 | MTA + Adhesive resin restoration |
| Case | Report | 20 ml of 6% NaOCl | 5 ml Saline | 10 ml of 2% CHX | the pulp chamber was sealed with a dentine bonding agent and flowable composite | No Instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 2 weeks and 4 weeks | Blood clot | MTA + Adhesive resin restoration |
|------|--------|------------------|-------------|---------------|-------------------------------------------------|-------------------|----------------------------------------|------------------|-----------|---------------------------------|
| 9    | Case report | 10 ml of 6% NaOCl + Saline + 10 ml of 2% CHX | The draining sinus tract was rinsed with 3 ml of 0.12% | Nothing used | # The draining sinus tract was rinsed with 3 ml of 0.12% CHX at the first visit and 2 weeks later and amoxicillin and ibuprofen was prescribed | In the same first visit | Blood clot | MTA + Adhesive resin restoration |
| 23   | Case series | 20 ml of 5.25% NaOCl | 20 ml Saline | 10 ml of 0.12% CHX | The saline rinse was used to prevent the interaction between NaOCl and CHX | No Instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 3 Weeks | Blood clot | MTA + Adhesive resin restoration CollaPlug is added in two cases |
| 18   | Case report | 30 ml of 1% NaOCl | No Instrumentation | Metronidazole + Ciprofloxacin + Amoxicillin | 6 weeks | Tooth as redressed at 3 weeks | Blood clot | MTA + Glass Ionomer + Amalgam |
| 19   | Case report | 5% NaOCl | 3% H2O2 | No Instrumentation | Calcipexin the upper part of the root canal | 6 weeks | Without inducing bleeding | Directly capping a vital tissue remained in the apical part of the root about 10 mm from the orifice | Vitapex + Glass Ionomer + Adhesive resin restoration |
| 17   | Case reports | 20 ml of 5.25% NaOCl for 20 minutes | Gates Glidden drills used passively in the coronal one third of the mesial canals | Metronidazole + Ciprofloxacin + Minocycline | 3 weeks | Blood clot | MTA + Glass Ionomer + Amalgam |
| 16   | Case Series | 10 ml of 2.5% NaOCl | 10 ml Saline | No Instrumentation | Ca(OH)2 in the coronal third of root | 3 weeks | Blood clot | MTA + Glass Ionomer + Adhesive resin restoration in 5 molars MTA + Glass Ionomer + Amalgam in 1 molar |
| Case/Report | Treatment | Instrumentation | Intravenous therapy | Duration | Additional Therapy | Outcome | Restoration |
|-------------|-----------|-----------------|---------------------|----------|-------------------|---------|-------------|
| 6, 7 Case report | 10 ml of 5.25% NaOCl | No instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 22 day | PRP | MTA + Cavit + Amalgam |
| 8 Case report | 10 ml of 3% NaOCl for 2 min or 20 ml of 3% NaOCl for 5 min + Saline | No instrumentation | Metronidazole + Ciprofloxacin + Cefaclor | 1 and 2 weeks | Blood clot | MTA + Obtura II + Adhesive resin restoration |
| 11 Case report | 6% NaOCl | No Instrumentation | Metronidazole + Ciprofloxacin + Minocycline + Thickening agent; carboxymethylcellulose | 3 weeks | | MTA + Adhesive resin restoration |
| 42 Case Series | 5% NaOCl + Saline | No instrumentation | Metronidazole + Ciprofloxacin + Minocycline + base of propylene glycol and Macrogol | 2-6 weeks | Blood clot | MTA + Glass Ionomer + Adhesive resin restoration |
| 28 Case Series | 5.25% NaOCl | instrumentation was minimal | Ca(OH)2 in the coronal half of root | 4 weeks | Blood clot Bleeding was not satisfactory in 6 cases | MTA + Adhesive resin restoration |
| 21 Case report | 10 ml of 2.5% NaOCl | No Instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 35 days | Blood clot | MTA + Adhesive resin restoration |
| 22 Case report | 10 ml of 2.5% NaOCl + 10 ml Saline | No Instrumentation | Ca(OH)2 | 3 weeks | Blood clot | MTA + Glass Ionomer + Adhesive resin restoration |
| 29 Case report | 5.25% NaOCl intermediated rinse of distilled water 2% CHX | Gentle instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 3 weeks | Blood clot | MTA + Glass Ionomer + Adhesive resin restoration |
| 10 Case report | 20 ml of 5.25% NaOCl + 10 ml of 0.2% CHX Saline | No instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 3 weeks | PRF | MTA + Glass Ionomer + Adhesive resin restoration |
| 30 A pilot Clinical study | 20 ml 2.5% NaOCl | Minimal instrumentation | Metronidazole + Ciprofloxacin + Minocycline | Not mentioned | | Glass Ionomer |
| 26 Case report | 20 ml of 5.25% NaOCl | No instrumentation | Metronidazole + Ciprofloxacin + Minocycline | 4 weeks | | MTA + Permanent restoration |

*Note: CHX stands for Chlorhexidine.*
### Table 5. Results

| Ref. | Follow Up Period | Radiographic Assessment | Sensitivity Testing | Other Observations | Complications |
|------|------------------|-------------------------|---------------------|-------------------|---------------|
| 1    | 30 months        | 1) Healing of periapical tissues  
                  2) Apical closure  
                  3) Root wall thickness increased | +ve EPT on the surface of newly formed dentinal bridge. | ___ | ___ |
| 27   | 24 months        | 1) Healing of periapical tissues  
                  2) Apical closure | inconclusive | ___ | ___ |
| 12, 13 | 12 months      | 1) Healing of periapical tissues  
                  2) Apical closure  
                  3) Root wall thickness increased  
                  4) Root length increased | -ve Cold and Electric pulp tests  
                  radiopacities in the canal space associated with partial pulp canal obliteration | ___ | ___ |
| 24   | 1-5 years        | 1) Healing of periapical tissues  
                  2) Apical closure  
                  3) Root wall thickness increased | ___ | ___ | ___ |
| 31   | 6 months-3 yrs   | 1) Healing of periapical tissues in 11 of the 14  
                  2) Root wall thickness increased in 8 of the 14  
                  3) Root length increased in 10 of the 14 |  | ___ | ___ |
| No | Duration | Details | Observations | Comments |
|----|----------|---------|--------------|----------|
| 35 | 12-15 months | In only 3 cases 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased | +ve response in 3 cases | 2 cases: pain during disinfection, 4 cases: failure to induce bleeding, 3 cases: dropout |
| 15 | 18 months | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased | +ve Cold pulp test | slight cervical discolouration possibly related to the use of grey MTA in one premolar And No discoloration in the other one |
| 9 | 19 months | 1) Healing of periapical tissues 2) Root maturation | -ve Cold and Electric pulp tests | complete resolution of condensing osteitis |
| 23 | 12 months | 1) Healing of periapical tissues 2) Apical closure Observed in 4 teeth out of 6 3) Root wall thickness increased in 4 teeth out of 6 | +ve Cold and Electric pulp tests in only 2 teeth out of 6 | The placement of MTA resulted in movement of the MTA further apically than desired. So CollaPlug was used in Case 2 and 3 |
| 18 | 18 months | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased | +ve Electric pulp test | No obvious change in color |
| 19 | 11 yrs | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased 4) Root length increased | +ve Electric pulp tests on the surface of newly formed dentine bridge | dentinal bridge was found subjacent to the filling material |
| 17 | 15 to 18 months | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased | | 1) Poor radiographic outcomes in mesial root in case 1 could partially be attributed to insufficient bleeding. 2) It is possible that the smaller mesial canal diameters with smaller apical openings that limit the use of larger files for irritating apical tissues are other reasons for this problem in both cases. |
| 16 | 10 months | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased 4) Root length increased | +ve Cold pulp test in 2 cases which did not instrumented before | The only study that shows the % of increase in root length and thickness |
| 6,7 | 14 months | 1) Healing of Periapical tissues 2) Apical closure 3) Root wall thickness increased 4) Root length increased | +ve Cold and Electric pulp tests | pain and sensitivity to cold after 14 month |
|   | Months | Response Details                                                                 | Satisfactory | Delayed | Failure |
|---|--------|---------------------------------------------------------------------------------|--------------|---------|---------|
| 11 | 24     | 1) Healing of periapical tissues                                                  | -ve Cold pulp test |         | Tooth discoloration |
| 28 | 6-26   | 1) Healing of periapical tissues in all teeth                                    |               |         |         |
| 22 | 18     | 1) Healing of periapical tissues                                                  | +ve Cold pulp test and Delayed response to Electric pulp test |         |         |
| 29 | 24     | 1) Healing of periapical tissues                                                  |               |         |         |
| 10 | 12     | 1) Healing of periapical tissues                                                  | +ve Cold and Electric pulp tests |         |         |

Five types of responses of immature permanent teeth with infected necrotic pulp tissue and either apical periodontitis or abscess to revascularization procedures were observed in the present case series:
- **Type 1**: There was increased thickening of the canal walls and continued root maturation in 15 cases in average of 19.6 months.
- **Type 2**: There was no significant continuation of root development and the root apex became blunt and closed in 5 cases.
- **Type 3**: There was continued root development and the apical foramen remained open in 5 cases.
- **Type 4**: There was severe calcification (obliteration) of the canal space in 4 cases in average of 16 months.
- **Type 5**: There was a hard tissue barrier formed in the canal space between the coronal MTA plug and the root apex in 2 cases.

The response of one incisor with successful apexification without revitalization of the root canal was unexpected, the most likely explanation is that the blood clot broke down, leaving an empty space with no scaffold into which the new vital tissue could grow.
| Age  | Follow-Up | Comments                                                                 |
|------|-----------|--------------------------------------------------------------------------|
| 30   | 12 months | 1) There was a statistically significant difference in periapical healing, apical closure, and dentinal wall thickening in PRP group compared with the other group. 2) Root lengthening was comparable in both groups. |
| 26   | 6 yrs     | 1) Healing of periapical tissues 2) Root apices formed without increase in length and thickness of the roots. -ve Cold pulp test. |
| 32   | 24 months | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased 4) Root length increased -ve Cold and Electric pulp tests. |
| 34   | 9 months  | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased -ve Cold and Electric pulp tests. |
| 14   | 24 months | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased -ve Cold pulp test. |
| 20   | 15 months | 1) Apical closure 2) Root wall thickness increased +ve Cold and Electric pulp tests. |
| 33   | 5 yrs     | 1) Healing of periapical tissues 2) Apical closure 3) Root wall thickness increased 4) Root length increased -ve Cold and Electric pulp tests. |
| 40   | 12 months | 1) Healing of periapical tissues 2) Apical closure -ve Cold and Electric pulp tests. |

The dis-advantages of this procedure include drawing blood in young patients, the need of special equipment and reagents to prepare PRP, and the increased cost of treatment. These are minor drawbacks when compared with improved healing and the regenerative response that was obtained with its use as documented in the present study.

Patient demands esthetic so root canal therapy, casting dowel core and full crown were planned. During root canal treatment, both root canals were empty, and no vital tissue or bleeding was seen.
Methodological steps: Table 4 is showing the methodological steps followed in the included studies during the regeneration procedures in terms of: study design, type of irrigant used, using instrumentation or not, used antibiotic paste, time lasts for controlling the infection, scaffold used and which material used for coronal sealing.

Results: Table 5 is showing the follow up period in each study, the radiographic findings in terms of; healing in the periapical radiolucency, apical closure and/or increase in root wall thickness. It also shows any other mentioned observations and the sensitivity test results if done.

7. Discussion

We are providing a narrative synthesis of the findings from the 31 included studies, structured around same PIO; P: Necrotic immature root, the differences were in the cause and history of necrosis and the clinical picture of the cases. I: Regenerative endodontic treatment but different methodological steps were followed; types of irrigants and antibiotics used to disinfect the root canal, variations in concentrations of the same used materials, number of visits required for treatment, scaffold used and materials used for coronal seal. O: Root development is the primary outcome together with resolution of signs and symptoms.

The available clinical cases all are published in form of case reports and case series. There is not any randomized controlled trial in this field. In the hierarchy of evidence, case series represents level IV evidence but they are the core in the review because they were the only available in-vivo clinical data.

The 31 included papers offer 115 treated teeth with regenerative endodontic treatments; premolars, incisors and molars, 37 females and 55 males with ages ranged from 8 to 28 years old. The clinical picture of the necrotic immature teeth varies between symptomatic apical periodontitis, asymptomatic apical periodontitis, acute abscess or chronic abscess. Pulp necrosis due to traumatic crown fractures luxates injuries, caries, and denst evaginatus and replanted avulsed tooth. These findings indicate that the procedure can be successfully planned in any tooth type and any other different situations.

To establish an applicable protocol for regenerative endodontic procedures, the influence of every step in the treatment techniques on the outcome has to be evaluated. These include case selection, methods used to control the infection, root canal irrigation and dentin pretreatment, root canal preparation and coronal sealing, timing of visits, and postoperative care. Additional aspects to support healing have to be considered, such as the choice of suitable scaffold materials, delivery or recruitment of a critical mass of stem cells, and the use of growth and differentiation factors.

Regeneration methods start with disinfection of the root canal, this step is considered the corner stone for the treatment success. The infection of the root canal system is polymicrobial infection. It is unlikely that any single antibiotic could result in effective sterilization of the canal. The common protocol used in regeneration utilizes different irrigants and antibiotic pastes with no or minimal instrumentation to create a sterile medium for the newly regenerated tissues.

Did not do instrumentation. Minimal or gentle instrumentation was done by. While did instrumentation in the coronal and middle thirds of the root using manual endodontic files and gates glidden drills. Frank instrumentation was done by as they enlarged the apical opening to 1.1 mm using 110 K file, while cleaned the canal mechanically but not to its full length.

The irrigants used varies between the reported cases; NaOCl, CHX, EDTA and Saline. They are used in different concentrations and combinations. NaOCl considered the main irrigant used in most of the articles but with different concentrations: 1%, 1.25%, 2.5%, 3%, 5%, 5.25%, 6%, 8%. CHX is a broad-spectrum antimicrobial agent and is effective against certain NaOCl resistant bacterial strains.

NaOCl is a tissue solvent and antimicrobial agent. It is toxic and can cause inflammation in the periapical tissues if used in high concentration, whereas it is ineffective against specific microorganisms in low concentrations. While CHX is a broad-spectrum antimicrobial agent and is effective against certain NaOCl resistant bacterial strains.

Not only should bactericidal/bacteriostatic properties of irrigants be considered but also their ability to promote survival and proliferative capacity of the patient’s stem cells. For that purpose studied the effect of some irrigants with different concentrations on stem cells and concluded that irrigation protocols that include 17% EDTA appear to promote SCAP (stem cells from the apical papilla) survival and attachment to the root canal dentinal wall. On the other hand, 2% CHX appeared detrimental to SCAP in the model used, yielding no viable cells. However, the addition of 6% NaOCl to EDTA decreased cell viability when compared with EDTA alone.
The combination of NaOCl and CHX results in the formation of a precipitate, its formation should be avoided by removing the NaOCl before placing CHX into the canal. Saline should be used between CHX and NaOCl to avoid any interaction between the irrigants.

Investigated the influence of irrigants on cellular behavior and differentiation. Surface conditioning with EDTA induced an intimate association of cells with dentin and differentiation into an od on to blast phenotype. Biocompatible irrigants are needed to promote stem cell attachment to root canal dentin, which is essential to accomplish some regenerative endodontic therapies.

Antibiotics used to complete the disinfection of the necrotic infected canals. Different mixtures are used but the commonly used is that of Metronidazole, Ciprofloxacin and Minocycline as used by while added a thickening agent “carboxymethylcellulose” to them. They replaced “minocyclin” with “cefaclor”, while replaced it with amoxicillin to avoid minocycin induced discoloration. Others used calcium hydroxide in the coronal third of root used calcium hydroxide and 2% chlorohexidine gel.

Calcium hydroxide is a well-known medicament for the asepsis of root canals because of its high alkalinity and antibacterial activity studied the cytotoxic effect of five intracanal medicaments on the viability of human stem cells of the apical papilla (SCAPs). The search concluded that (ciprofloxacin, metronidazole and minocycline), (ciprofloxacin and metronidazole), (ciprofloxacin, metronidazole and cefaclor) and Augmentin significantly reduced SCAP survival at high concentrations while calcium hydroxide was conducive with SCAP survival. did not use any antibiotic pastes but used formocresol as an inter appointment dressing together with irrigants as well did not use antibiotic pastes but rinsed the draining sinus tract with 3 ml of 0.12% chlorhexidine gluconate at the first visit and 2 weeks later and amoxicillin and ibuprofen was prescribed.

Once the infection control is achieved through resolution of the clinical signs and symptoms, the regeneration procedures are followed and completed in single visit. Regarding the need for scaffold as a matrix for the newly formed tissue; a blood clot formation inside the canal is considered a scaffold and a source of growth factors. Inducing bleeding to promote blood clot formation was the commonly used method in most of the reported cases added an absorbable scaffold to the induced bleeding. While did not induce bleeding as there seemed to be some vital tissues remaining in the apical half of the canal as they mentioned that insertion of an endodontic file evoked a sensation of pain used concentrated platelet rich plasma as a scaffold and autologous supply of growth factors used platelet rich fibrin for the same purpose.

A tight coronal seal is placed to prevent ingress of any microorganisms. MTA was the material of choice in all of the reported cases except for who used glass ionomer and as for who used calcium hydroxide; then the access cavity was sealed with glass ionomer, adhesive resin restoration or amalgam.

Analyzing the radiographic results by examining the status of the periapical tissues and changes in the apical part of the root, root walls and root length. Results reveal that all cases showed healing of periapical tissues and apical closure and these are promising results to encourage generalization of the treatment in the daily practice.

Reported complications are pain and sensitivity after 14 months from therapy, tooth discoloration or not enough bleeding.

To avoid discoloration; avoid minocycline used white MTA or sealed the pulp chamber with a dentine bonding agent and flow able composite before placement of the antibiotic paste.

To get an evidence based guidelines, further research is needed pertaining to the various disinfectants, scaffolds, growth factors and coronal sealing materials that can be used for dental pulp-tissue engineering. Since advantages of regeneration is that; it can be completed in a single visit once the infection is controlled, so there are no need for multiple appointments; as in the case of calcium hydroxide apexification. It is also very cost-effective. The biggest advantage is that achieving continued root development and strengthening of the root as a result of thickening of the lateral dentinal walls.

8. References

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