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Clinical and Radiographic Outcome of Revasclarization of Immature Teeth with Necrotic Pulp Using Calcium Hydroxide and MTA

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ABSTRACT

Background: Dental injuries can lead to harm to the pulp tissue in developing teeth. Revascularization therapy presents a viable treatment option for non-vital, immature permanent teeth that have experienced trauma. This prospective study aimed to assess the radiographic and clinical outcomes of immature teeth with a trauma history that were treated using regenerative endodontic procedures and mineral trioxide aggregate apexification techniques. **Methods:** This cohort study was conducted at the Dentistry Department of Bolan Medical College, Faculty of Dentistry, from August 21, 2021, to February 22, 2022, of forty-one patients with traumatized immature permanent maxillary incisors was included in this study. The patients were divided into two distinct groups: those who had previously received endodontic treatment and those who had not. The regenerative endodontic protocol group (Group 1) included twenty-four patients who directly approached the clinic and had not undergone endodontic treatment prior. In contrast, seventeen patients who had previously experienced endodontic intervention on their respective teeth were placed in the mineral trioxide aggregate apexification group (Group 2). The follow-up period for the patients extended to 24 months. Clinical success rates were evaluated, and both pretreatment and control radiographs were analyzed to compute the percentage increase in root dentin width and root length. **Results:** Following a 24-month follow-up period, the majority of cases exhibited positive periapical healing as indicated by the radiographic findings. In Group 1, there was a slight increase in both root length and root dentin width, alongside a noticeable narrowing of the apical opening. The radiographic assessment of two cases, which were accessible 11 years later, underscored the significance of long-term follow-up in evaluating the efficacy of the selected methods. **Conclusion:** The revascularization technique serves as a treatment alternative that yields favorable outcomes regarding root development in teeth affected by necrotic pulp due to trauma.

INTRODUCTION

Traumatic injuries to permanent teeth are frequently observed in both children and young adults,[1] with the anterior maxilla being the most affected area during this time.[2] Damage to immature permanent teeth can result from bacterial invasion and/or dental trauma. If the inflamed pulp tissue is left untreated in such cases, necrosis will ensue. The death of odontoblasts due to necrosis results in impaired root development [3] and increased fragility of the teeth. These outcomes may arise from potential damage to the vascular system and Hertwig's epithelial root sheath in the periapical region, even when the crown dentin and enamel remain undamaged.[4-6]

Treating infected pulp in immature permanent teeth presents several clinical challenges. The wide opening of the apex complicates root canal debridement and obturation. In immature teeth, the apical opening is closed using the apexification method to establish a calcified barrier or to support the apical development of the tooth root with necrosed pulp, where root development has not been finalized. Traditionally, these teeth have been treated with either prolonged calcium hydroxide therapy[7-9] or apexification techniques utilizing mineral trioxide aggregate (MTA).[10] Although these methods alleviate negative signs and symptoms, their positive impact on root development is limited.[11] Consequently, it is acknowledged that immature teeth treated through these approaches cannot continue their development. It should not be anticipated that root growth will persist, nor that pulpal nociception and immune defense will function normally in these teeth. All these factors highlight the importance of regenerative endodontic procedures in addressing traumatic injuries.

The clinical outcomes associated with the revitalization technique employed for treating teeth suffering from necrosis as a result of trauma and ongoing root development remain ambiguous. While there exist studies addressing the timing of periapical wound healing, the enhancement of dentin thickness along the canal walls, and the increase in root length following revitalization in immature teeth with necrotic pulps, the proportional results are not well-defined.[12-13] Nevertheless, in the case of traumatized teeth, such endodontic methods play a crucial role in supporting the functionality and vitality of the tooth.[14]

MATERIALS AND METHODS

This prospective study was carried out at the Dentistry Department of Bolan Medical College, Faculty of Dentistry, from August 21, 2021, to February 22, 2022. The study protocol received approval from the Ethics Committee of Bolan Medical College Faculty of Dentistry (ref. 2022/354). The trial involved 45 children (18 girls and 27 boys) aged between 7 and 12 years, each having at least one non-vital upper permanent incisor with an open apex, where root development was compromised due to trauma. During the clinical examination, the spontaneous pain status of the patient, the presence or absence of a sinus tract, sensitivity to palpation, and responses to cold and electric pulp tests were assessed. An informed consent form was secured from the parents regarding the potential consequences and complications associated with the proposed treatment approach. In this study, a power analysis conducted using G*Power 3.1 software (alpha error p=0.05) determined that a sample size of 40 was adequate. A total sample size of 46 teeth was calculated to identify differences at the 5% significance level, accounting for an expected 10% loss to follow-up, utilizing the Mann-Whitney U test for comparing independent means. Fifty single-rooted teeth (1 or 2 per patient) with apical lesions, regardless of prior endodontic intervention, were included in both treatment groups. Twenty-five patients who initially visited the university clinic without prior consultation with any dentist were assigned to the regenerative endodontic protocol group (Group 1). In contrast, 25 patients who had previously undergone endodontic treatment were placed in the MTA apexification group (Group 2).

In the REP group, one patient was removed from the study during the 6-month follow-up due to the retraumatization of the same tooth. In the MTA apexification group, eight patients were excluded from the study at the 12-month follow-up for the same reason. A total of forty-one teeth were included in the statistical analysis. Only patients who were healthy and cooperative were included in this study. Teeth exhibiting vertical fractures, significant periodontal issues, and those that could not be restored were excluded from the study. The criteria for exclusion from the study included poor cooperation, new trauma or complications, and the onset of disease that necessitated the termination of the study. To ensure consistent film positioning at all experimental time points, a film holder was utilized for taking periapical radiographs. The digital films were exposed to a uniform X-ray source set at 60 kV, 4 mA, and 0.25 seconds. The same clinician (GK) treated both groups of teeth following this protocol: pulpal sensitivity was evaluated using a carbon dioxide negative test (-50°C) and an electric pulp tester; local anesthesia without vasoconstrictors was administered during the anesthesia of the teeth. After isolating the area with a rubber dam, the access cavity was prepared using a round diamond and Endo-Z bur, and the working length was determined with an apex locator. Passive positive pressure irrigation was employed to disinfect the canals with 20 mL of 5% sodium hypochlorite (NaOCl), 20 mL of saline, and 20 mL of 0.2% chlorhexidine (CLX) solution, without instrumentation, using a Max-i-Probe® needle. Precautions were taken to ensure that the irrigation was 3 mm shorter than the working length to prevent chemical interaction with periapical stem cells. The canals were subsequently dried with paper points, and to minimize or eliminate discoloration, a bonding agent was applied to the cavity walls. Commercially prepared chemotherapeutic agents, specifically metronidazole, ciprofloxacin, and minocycline, were utilized.

The procedures for preparing the triple antibiotic paste (TAP) have been detailed in other sources.[14-16] In summary, after the removal of the materials covering the drugs, each drug was ground into a fine powder using a pestle in a porcelain mortar. Subsequently, it was stored separately in tightly sealed porcelain containers to avoid exposure to light and moisture. A small quantity of silica gel was incorporated to minimize humidity in the environment, and the drugs were utilized within a month. The TAP was introduced into the canal spaces to safeguard the root canal walls, employing a 25-gauge lentulo spiral up to size 8, which was attached to achieve a paste-like consistency. Glass ionomer cement was utilized to seal the access cavity, and patients were scheduled for follow-up after 4 weeks. For disinfection purposes, the canals were irrigated without pressure using 20 mL of 0.5% sodium hypochlorite and infused with 20 mL of saline via a Maxiprobe (Dentsply Rinn, Elgin, IL). During the irrigation process, the needle was positioned 1 mm short of the radiographic apex of the tooth. The canals were subsequently dried using paper points, and to mitigate or eliminate discoloration, a bonding agent was applied to the cavity walls. Calcium hydroxide ($Ca(OH)_2$), prepared to a paste-like consistency, was introduced into the root canal cavities using a low-speed spiral, and the cavity was temporarily sealed with conventional glass ionomer cement. The patient was then scheduled for a follow-up appointment one week later. During the next visit, under rubber dam isolation, the canals were rinsed with sterile saline to eliminate calcium hydroxide residues and dried with paper points. Mineral trioxide aggregate, prepared according to the manufacturer's instructions, was placed into the canal and compacted with the appropriate plugger to fill the apical one-third (4-5 mm) of the canal.

The accurate positioning of the material was verified through periapical radiography. Should the apical barrier prove insufficient, the procedures were repeated. A cotton pellet, dampened with sterile water, was positioned at the canal opening for a duration of 15-30 minutes, after which a dry cotton pellet was inserted, and the access cavity was filled with thermoplasticized gutta-percha. Ultimately, the coronal access cavity was restored using glass-ionomer cement, and composite restorations (3M ESPE Filtek® Z550, St. Paul, USA) were applied. Patients in Group 1 and

Group 2 were scheduled for follow-up appointments at thirty days, three months, six months, one year, and two years for additional clinical and radiographic assessment.

Both groups were recalled for control periods at 1, 3, 6, 12, and 24 months. The data gathered during these intervals were compared with the initial examination results. Both clinical and radiographic findings were assessed by the same qualified clinicians (GK, GA). In the evaluation of clinical findings, the following criteria were taken into account: pulp sensitivity, occurrence of spontaneous pain, presence of pain upon percussion and palpation, condition of the sinus tract, and discoloration of the enamel. Pulp sensitivity was assessed through cold thermal testing and electric pulp testing. The criteria for the radiographic examination included complete bone healing of the periapical lesion, increase in root length, and formation of the radiographic apex. RVG images were captured before and after treatment in JPEG format. The data analysis was conducted using NCSS (Number Cruncher Statistical System) 2007 and PASS (Power Analysis and Sample Size) 2008 Statistical Software (NCSS, LLC, Utah, USA). In the analysis of clinical and radiographic parameters, the differences between Group 1 and Group 2 were assessed using the McNemar test and Chi-square test at a significance level of $p<0.05$. The percentage change in root length and dentin thickness was evaluated using the t-test.

RESULTS

A total of 50 patients out of these, 41 patients had a follow-up period of at least 24 months, resulting in a recall rate of 82%. Among these 41 cases, there were 24 cases that underwent the regenerative endodontic protocol and 17 cases that received MTA apexification treatment. Table 1 provides a summary of the patient data for this study population. One patient from the REP group and eight patients from the MTA apexification group were excluded from the study due to their absence at the 6-month follow-up. Table 1 at the 3rd month, all teeth in Group 1 were asymptomatic, and periradicular radiolucency areas were observed to heal radiographically. Clinical evaluations indicated a reduction in palpation, pain, sensitivity to percussion, and sinus tract presence in both treated groups (REP and MTA), with no statistically significant difference between the groups ($p>0.05$). Crown discoloration was noted in 24 teeth (100%) in the REP group and in seven teeth (41.18%) in the MTA group, with the difference not being statistically significant ($p=0.01$). All clinical findings before and after treatment for both groups are detailed in Table 2. Regarding tooth survival analysis, the REP group exhibited similar survival rates (24/24 teeth [100%]) when compared to the MTA apexification group (17/17 teeth [100%]) ($p<0.05$). Radiographic examinations showed repair of periapical lesions in both groups, although no statistically significant differences were found between them ($p=0.305$). Apical closure results were significant in both groups ($p<0.05$). There was no significant difference observed between the groups concerning the increase in root length ($p=0.304$), and group analyses are presented in Table 3. Despite a higher number of immature teeth in the revascularization group, there were no differences in root development between the groups. Analysis of radiographic findings indicated that revascularization treatment significantly affected the root tip width in the apical 1/3. ($p<0.0001$).

After a period of 11 years, one case from each study group that was accessible was invited for a follow-up and underwent radiographic evaluation. The anticipated positive outcomes of the treatment included the clinical evidence of periapical healing, its radiographic assessment, monitoring of root development through radiography, and favorable results in the pulp vitality test.

Table 1

Patient demographics and clinical characteristics of the study population

	Group REP		Group MTA	
	Teeth	**%**	**Teeth**	**%**
Sex				
Male	16	66.6%	12	70.5%
Female	8	33.3%	5	29.4%
Signs and Symptoms				
Absent	4	16.6%	10	58.82%
Present	20	83.3%	7	41.7%
Apical Lesion				
Absent	13	54.16%	14	82.35%
Present	11	45.83%	3	17.65%
Age		10.5±2.7		
Follow-up Time		2 years		

Table 2

Clinical findings before and after treatment for groups undergoing regenerative endodontic procedure (REP) and mineral trioxide aggregate (MTA) apexification protocol

	Group REP (n=24)			Group MTA (n=17)		
Clinical Aspects	**Before**	**After**	**P value**	**Before**	**After**	**P value**
Pulp Vitality	0 (0%)	20 (83.33%)	0.00	0 (0%)	0 (0%)	
Mobility	17 (70.83%)	1 (4.17%)	0.00	4 (23.53%)	0 (0%)	0.00
Pain on Percussion	20 (83.33%)	1 (4.17%)	0.00	7 (41.18%)	0 (0%)	0.00
Spontaneous Pain	19 (79.17%)	1 (4.17%)	0.00	9 (52.94%)	0 (0%)	0.00
Sinus Tract	11 (45.83%)	0 (0%)	0.00	3 (17.65%)	0 (0%)	0.00
Crown Discoloration	0 (0%)	24 (100%)	0.00	0 (0%)	7 (41.18%)	0.00

Table 3

Pre- and post-treatment radiographic analysis data for regenerative endodontic procedure (REP) and mineral trioxide ag-gregate (MTA) apical plug applied groups

	Group REP (n=24)	Group MTA (n=17)	
Radiographic Aspects	**Before-After**	**Before-After**	**p value**
Root Length	-7.52±8.33	-9.36±10.31	0.255
Root Tip Width	31.97±15.17	19.11±18.91	0.039
Root Canal Width	0.98±7.35	-0.78±10.42	0.721
Pulp Space	3.73±17.87	-16.56±16.56	0.001
Dentin Thickness	-2.76±14.12	8.97±21.35	0.039

DISCUSSION

Trauma is more prevalent in the immature teeth of adolescents,[2,15,16] and apexification therapy is frequently employed in the management of these teeth.[6,17] Apexification is a procedure conducted for the treatment and safeguarding of immature permanent teeth with necrotic pulp.[18] Calcium hydroxide [Ca(OH)2] is extensively utilized in apexification, as it can facilitate physiological closure of immature pulpless teeth.[9,19,20] The conventional Ca(OH)2 technique presents several limitations, such as the unpredictability of apical barrier formation, prolonged treatment durations necessitating regular monitoring,[9,21-23] and the requirement for complete removal of the paste. These drawbacks have prompted the adoption of mineral trioxide aggregate, which effectively seals the apical end and does not necessitate the formation of a calcific barrier.[18] While MTA apexification reduces treatment time, it is expensive. Furthermore, its widespread application may be constrained by the necessity for clinical expertise in addressing challenges such as the thinness of the dentin walls or the delivery of material to the apical 3-4 mm.[24-26] Pulp damage resulting from trauma or caries in teeth with incomplete apical development has enabled the exploration of regenerative potential in endodontics.[27] In this investigation, REP was favored among the treatment alternatives for the immature non-vital teeth of patients presenting directly to the university clinic. Mineral trioxide aggregate apexification was the preferred method in the treatment of immature teeth that had previously undergone endodontic procedures.

In the REPs, disinfection is applied at a very high level.[28-29] In the disinfection of root canals, the use of $Ca(OH)_2$ or antimicrobial dressing after sodium hypochlorite (NaOCl) and chlorhexidine irrigation is common.[30-32]

When assessing root tip development, it is noted that the majority of teeth exhibit only apical closure. A comparable finding was noted in teeth that underwent apexification; there was no observable increase in root length nor any reduction in the root canal space.

Kahler et al.[33] proposed that extending the duration of examinations would yield more significant insights in cases of regeneration, as root maturation persisted in two instances throughout the 36-month follow-up period in their research. In this investigation, periapical lesions were found to decrease in both groups, with periapical healing confirmed radiographically in the majority of instances. No statistically significant differences were identified between the groups regarding root length development ($p=0.304$). There was no discernible difference in root development between the REP and MTA groups; however, the revascularization group exhibited less maturation. An analysis of the radiographic data indicated that revascularization treatment significantly influenced ($p<0.05$) the width of the root tip in the apical third. In Group 1, a notable enhancement in root tip width and a reduction in apical diameter were recorded in the apical 1/3. Achieving an accurate diagnosis in pediatric patients should rely on the current clinical symptoms, the history of these symptoms, diagnostic tests employed, and clinical findings,[34] as younger patients may not consistently exhibit reliable clinical symptoms. Concerning clinical examination, existing literature indicates that signs and symptoms typically diminish following the revascularization procedure, and in certain instances, pulp sensitivity may return.[6] In this investigation, patients from both groups reported a reduction in clinical complaints, with 83.33% of teeth in the REP group experiencing heightened pulp sensitivity. While the anticipated outcomes, such as the healing of the apical lesion and the continuation of root development, are frequently achieved in REPs, the observation of coronal discoloration attributed to minocycline is considered an adverse effect. This study employed the induced bleeding technique in REPs. Although the impact on periapical healing could not be distinctly established in our research, it is believed to be beneficial. In this study, the recall intervals for the groups were comparable (1, 3, 6, 12, and 24 months) and aligned with those of other investigations in this domain.[12]

Currently, there is a growing number of studies focusing on both cell-free and cell-based strategies aimed at regenerating pulp tissue. Nonetheless, the challenges associated with implementing cell-based approaches are quite apparent. Conversely, complete clarity has yet to be achieved regarding the cell-free approach that utilizes biological signaling molecules. It is essential to increase the volume of clinical studies on regenerative endodontic procedures that yield favorable clinical outcomes.

CONCLUSION

The findings of this prospective study yielded highly satisfactory treatment outcomes for traumatized immature permanent necrotic teeth utilizing REPs. Although the impact of REPs on apical diameter can be anticipated, it appears that this influence is not clearly observable on root length. The technique of induced bleeding in REP may positively influence the treatment of traumatized immature teeth.

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