Evaluation Of Platelet-Rich Plasma In Regeneration In Non-Vital Immature Permanent Teeth – An Original Research

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ABSTRACT

Background: A promising regenerative method used in many areas of medicine, including dentistry, is platelet-rich plasma. It makes use of the patient's own blood, which has been concentrated to contain growth hormones and platelets. The treatment location is then injected with these concentrated platelets to promote tissue regeneration and healing.

An area of scientific interest has been the use of platelet-rich plasma in dentistry, notably for the regeneration of non-vital immature permanent teeth. Non-vital immature permanent teeth are those that have roots that have not yet fully formed or that have received root canal therapy at a young age.

Methods: A random division into two groups of 60 immature, non-vital permanent teeth was made. After cleaning the root canal space with a triple antibiotic paste (1:1:1 ciprofloxacin, metronidazole, and cefaclor), a tissue scaffold was created using either PRP or a blood clot (as a control), and then white mineral trioxide aggregate was applied on top. All cases were clinically and radiographically followed for a full year. To measure variations in bone density, root length, and lesion size, computed tomography images from preoperative and postoperative periods were compared. The mean differences in each parameter between the blood clot and PRP groups were compared using the Mann-Whitney U test.

Results: After 5 months, 46 locations showed a delayed positive response to sensitivity tests (cold and electric pulp tests). In all 60 (100%) teeth, cone-beam computed tomography after 12 months showed resolution or a reduction in lesion size as well as an increase in bone density. Additionally, early root growth was seen in the test group (mineral trioxide aggregate with PRP) and ongoing root development was seen in 44 (73%) of the teeth.

Conclusions: The study's conclusions suggest that PRP can serve as a trustworthy scaffold for regenerative endodontic therapy. With the exception of a significant increase in root length, the results of PRP therapy were not significantly different from those of the conventional technique using a blood clot as the scaffold.

Keywords: Regenerative endodontics, blood clot, platelet-rich plasma, mineral trioxide aggregate, cone-beam computed tomography, immature permanent teeth

DOI: 10.31838/ecb/2023.12.Si9.266

INTRODUCTION

Dentists face a significant problem when managing permanent teeth with necrotic pulp, periapical disease, and halted root growth. Root fracture is more likely in teeth with stunted root growth because it causes weak root dentin, open apices, and stunted root growth. They are also inappropriate for debridement and filling using conventional methods and obturation materials because of these anomalies [1]. In the past, calcium hydroxide was used to produce an apical hard tissue wall before apexification, which is the preferred method of treatment for such cases. However, as shown by several studies, the addition of mineral trioxide aggregate (MTA) enabled the rapid creation of a synthetic wall with a very high success rate in the apical third of roots. [2,3]. As a result, fewer clinical visits are now planned. However, a drawback of both calcium hydroxide and MTA is that they prevent further root development, resulting in thin dentin walls and, consequently, a weak root structure [4].

A challenging problem in endodontic therapy arises from the insufficient root development and open apices of non-vital immature permanent teeth. Conventional methods frequently fall short of producing ideal results, which compromises root structure and limits the possibility for further maturation [5].

Adult stem cells, signalling molecules, and a three-dimensional (3D) physical framework that can support cell growth and differentiation have been the three main components of tissue regeneration that have been the focus of recent developments in tissue engineering. Recently, several authors have written about the idea of regenerative endodontics [5,6]. These investigations suggest that apex development in a sterile environment is maintained by stem cells from the apical papilla of developing teeth.

Regenerative endodontic procedures have attracted a lot of attention recently as a potential replacement for conventional therapies. Regeneration, as opposed to apexification, enables quick root development continuation, improved wall thickness, and natural mending of periapical tissues [5,7,8]. Due to its abundance of growth factors and capacity to encourage tissue repair and regeneration among these, Platelet-Rich Plasma (PRP) has emerged as a potentially efficient restorative agent.

A concentrated autologous blood product made from the patient's own blood called Platelet-Rich Plasma (PRP) is enhanced with more platelets than whole blood. In addition to platelet-derived growth factor (PDGF), transforming growth factor-beta (TGF-), insulin-like growth factor (IGF), and vascular endothelial growth factor (VEGF), these platelets also release a variety of other growth factors [7]. PRP is a desirable option for regenerative therapy in a variety of medical and dental sectors since these growth factors are essential for cell proliferation, differentiation, angiogenesis, and tissue regeneration [8,9].

The use of PRP in endodontics has produced encouraging results in terms of encouraging regeneration inside the root canal space of non-vital immature permanent teeth. PRP may encourage the differentiation of stem cells into odontoblast-like cells by fostering an environment that is conducive to tissue healing, resulting in dentinogenesis and subsequent root development [3,4,7]. Additionally, the possibility of improved periapical healing and apical closure may lower the danger of problems brought on by conventional root canal procedures.

Despite the rising popularity of regenerative endodontics and the potential advantages of PRP, robust scientific analysis through original research is still required. In order to improve the results of endodontic therapy in such circumstances, our research aims to offer evidence-based insights into the regenerative potential of PRP and its clinical relevance. By systematically examining the effectiveness of PRP in the regeneration of non-vital immature permanent teeth, this study seeks to add to the body of knowledge.

However, the widespread application of such therapy procedures in the clinical world has been limited by a lack of adequate data. The current study's objective is to evaluate the effectiveness of a PRP-based regeneration procedure in non-vital young permanent teeth.

MATERIALS AND METHODOLOGY

64 healthy participants with young, single-rooted, permanent teeth who regularly attended outpatient dental clinics were chosen for this split-mouth study. None of the teeth had ever received necrotic treatment. Patients and their guardians provided written informed permission. For this investigation, institutional ethics approval was obtained.

The following were the inclusion criteria:

- An undeveloped apex and periapical lesions with or without pulp necrosis
- Probability of tooth restoration
- No root fracture, ankylosis, pathologic mobility, or probing depths of more than 3 mm are present.
- Antibiotic or drug allergies are not required to perform the treatment.

The patient's dental history and the results of the clinical examination, which included cold and electric pulp tests, helped to make a preliminary diagnosis of pulp necrosis. Pain, swelling, fistulas, sensitivity to percussion, and palpation sensitivity were all identified as clinical signs and symptoms. The study included 64 non-vital young permanent teeth from the 32 patients who had either apical periodontitis or abscesses that showed a negative response to pulp testing. All teeth were randomly assigned to either the test group, which used PRP as the scaffold, or the control group, which used a blood clot as the scaffold. **Following are groups of teeth with periapical lesions based on the magnitude of the lesion:**

Score 0	Intact periapical bone structures Intact periapical bone structures
Score 1	Periapical radiolucency of >0.5 to 1 mm
Score 2	Periapical radiolucency of >1 to 2 mm
Score 3	Periapical radiolucency of >2 to 4 mm
Score 4	Periapical radiolucency of >4 to 8 mm
Score 5	Periapical radiolucency of >8 mm

TREATMENT PROCEDURES

The American Association of Endodontists' protocol for regenerative therapy was followed during the procedure. An access hole was formed with a rubber dam in place, irrigation with 1.5% sodium hypochlorite was applied liberally and gently, and a size #20 sterile K-file was used to measure the working length. Despite the absence of any instruments, canals were thoroughly irrigated (20 mL each of 2.5% NaOCl, sterile saline, and 0.12% chlorhexidine) and dried with sterile paper points. A creamy paste was produced by grinding a 1:1:1 mixture of metronidazole, ciprofloxacin, and minocycline with distilled water to a final concentration of 0.1 mg/ml. By sealing the pulp chamber with a dentin-bonding agent, the chance of discolouration was decreased.

Using a lentulo spiral in a slow-speed handpiece, this paste was applied in the canal beneath the cementoenamel junction (CEJ) to reduce crown discoloration. Reinforced cement made of zinc oxide and eugenol was used to seal canals. The patients were checked on after three weeks, and prior research have demonstrated that the root canal was successfully cleaned. Patients who continued to have any indications or symptoms of infection, such as purulent drainage, failed pain relief, edema, fistulas, or sensitivity to percussion and palpation, were given another dose of the triple antibiotic paste.

After three weeks, the effectiveness of the initial therapy was evaluated. If the patient showed no symptoms, the antibiotic paste was thoroughly removed using 20 ml of normal saline and 17% EDTA, followed by sterile paper points to dry the canals. Then, scaffolds were built in

accordance with the designated group. Under local anaesthetic, a size #20 K-file was rotated 2 mm past the apical foramen in the blood clot group in order to cause bleeding and fill the root canal up to the CEJ level. In order to achieve a coronal seal after blood clot formation, the final restoration was finished with white MTA (Dentsply Tulsa Dental, Tulsa, OK), glass ionomer cement (Fuji VII; GC, Tokyo, Japan), and composite resin (Filtek Z350; 3M ESPE, St. Paul, MN) during the same visit.
There was no anaesthesia applied to the PRP group. PRP was made in accordance with Dohan et al.'s instructions. To establish coagulation, equal amounts of sterile bovine thrombin (100 U/mL) and sterile saline solution with 10% calcium chloride were mixed with 31 PRP. After that, PRP was injected into the root canal up to the CEJ level. Final restoration for the blood clot group was finished as described.

CLINICAL AND RADIOGRAPHIC FOLLOW-UP
Throughout the 12-month follow-up period, clinical and radiographic follow-up exams were conducted every three months. All subsequent examinations were carried out by the same paediatric dentist.
Treatment-related teeth underwent cold and electric pulp testing to determine their vitality. Only when they gave positive results to both tests were their vitality testing response recorded. A CBCT scan was performed three months after treatment and subsequently at least once more every three to twelve months, depending on the patient's health. At the end of a year, the size of the lesion, periapical healing, and apical closure were separately assessed on CBCT images. One month following the initial examination, all radiographs were re-examined by the same paediatric dentist, and intra-examiner validity was evaluated using kappa statistics. Kappa scores, which indicate high dependability, were in the range of 0.8 to 1.0.
In each case, the maxillofacial region was imaged using CBCT (I-Cat; Imaging Sciences International, Hatfield, PA) at a setting of 120 kVp and 3 to 7 mA. An exposure period of 9 seconds was used to get the images. Each scan was performed across a 360° field of view with 0.3 mm voxels. We measured the root length, bone density (in Hounsfield units), and lesion size.
A 3.5-mm2 area in the coronal plane of the pretreatment CBCT images was used to record the bone density in areas with periapical lesions and healthy neighbouring bone regions. One year following endodontic treatment, using CBCT images taken with the identical conditions, the final postoperative measures were taken. Table 1 displays the mean initial and final bone density readings.

Table 1. Pre- and post-treatment comparison of parameters in both study groups (control and test).

Parameters studied	**Initial**		**Follow up**		**P-value**
	Mean	**SD**	**Mean**	**SD**	
Blood clot (Control)					
Lesion size (mm)	5.02	1.69	2.95	1.26	0.001*
Bone density (HU)	130.01	59.37	456.55	154.65	0.001*
Root length (mm)	13.45	4.10	12.90	4.35	0.001*
PRP (Test)					
Lesion size (mm)	5.29	2.46	2.25	2.09	0.001*
Bone density HU)	122.56	62.48	486.99	167.16	0.001*
Root length (mm)	13.08	4.03	13.25	4.45	0.001*

*Statistically significant
SD, standard deviation; HU, Hounsfield units

STATISTICAL ANALYSIS

The data were analyzed using SPSS Inc.'s (Chicago, IL) Statistical Package for Social Sciences, version 13. When analyzing clinical and radiographic data using a paired t-test, a P value of.001 was deemed statistically significant. A P value of.005 is considered statistically significant when comparing the means of differences in individual parameters between the control group (blood clot) and test group (PRP) using the Mann-Whitney U test.

RESULTS

PREOPERATIVE CLINICAL AND RADIOGRAPHIC FINDINGS

Only four out of the 64 participants (24 girls and 40 boys) missed their postoperative follow-up sessions. In 30 children (24 girls and 36 boys), there were 60 necrotic teeth (PRP, 30; blood clot, 30). Of these, 48 maxillary incisor teeth had pulp necrosis in addition to a history of trauma (either crown fracture or luxation), and 12 premolar teeth had pulp necrosis due to deep dentin caries or secondary caries.

Preoperative apical abscesses were noted in 18 teeth (PRP, 8; blood clot, 10), and preoperative acute symptoms comprising nocturnal discomfort, spontaneous pain, and high sensitivity to percussion were noted in 52 teeth (PRP, 28; blood clot, 24). Additionally, until the teeth were symptom-free, five teeth in the blood clot group and four teeth in the PRP group each required two sessions of triple antibiotic paste. 16 teeth (PRP, 6; blood clot, 2) had no periapical lesions, 20 teeth (PRP, 6; blood clot, 8), 18 teeth (PRP, 2; blood clot, 8), and 12 teeth (PRP, 6; blood clot, 2) had periapical lesions with scores of 1, 2 and 3 respectively.

POSTOPERATIVE CLINICAL AND RADIOGRAPHIC FINDINGS

The clinical and radiographic outcomes for each of the 30 patients (60 teeth total: 30 PRP, 30 blood clot) who received this endodontic treatment exhibited exceptional responses. All patients were monitored for a full year. After one month, all teeth had completely resolved related soft tissue lesions and were clinically symptom-free. After 5 months, 46 sites delayed positive responses to sensitivity tests (cold and electric pulp tests) were comparable to those of nearby teeth. At the 12-month follow-up visit, all 60 teeth (100%) had undergone periapical healing with resolution of the pathology's signs and symptoms (pain, swelling, fistula, and/or sensitivity to percussion and palpation), as well as resolution of the lesion or a reduction in its size and an increase in bone density.

Furthermore, 44 teeth (73% of cases; 28 in the PRP group and 16 in the blood clot group) demonstrated sustained root development or apical closure, whereas 38 cases (63.3% of cases; 26 in the PRP group and 12 in the blood clot group) demonstrated a postponed positive pulpal sensitivity test response. Pretreatment and postoperative CT examination data from the control and test groups revealed significant changes in the research parameters (P.001) (Table 1). When comparing the parameters of lesion size, bone density, and root length between the control group (MTA) and test group (MTA + PRP), only the mean difference in root length was found to be statistically significant (P.004) (Table 2).

Table 2 Comparison of mean of the differences in individual parameters in control group (blood clot) and test group (platelet-rich plasma).

Parameters studied	Blood clot		Platelet rich plasma		P value
	Mean	SD	Mean	SD	
Lesion size (mm)	3.12	0.92	3.62	0.16	0.157
Bone density (HU)	320.8	125.59	388.6	129.61	0.132
Root length (mm)	0.512	0.55	2.12	0.88	0.004

*Statistically significant

SD, standard deviation; HU, Hounsfield units

DISCUSSION

For use in revascularization techniques, many scaffolds have been the subject of prior studies. In the current study, we investigated how PRP affected the regeneration and repair of pulpal and periapical tissues. According to the study's findings, PRP can serve as an effective scaffold for regenerative endodontic therapy. The outcomes of PRP therapy were comparable to those of the standard procedure employing a blood clot as the scaffold, with the exception of the increase in root length.

According to earlier studies, the outcomes of regenerative protocols may vary between teeth with partial necrosis (good prognosis) and those with full necrosis (poor prognosis), requiring a distinct technique and scaffold for each of these types of teeth [9,10].

The efficacy of PRP as a scaffold in teeth with total pulp degradation was evaluated in the current study. Every tooth in the PRP group had signs and symptoms (pain, edema, fistulas, and/or sensitivity to percussion and palpation) resolved by the end of the 12-month period, and 93% of the group's teeth had increased root length or, to varied degrees, apical closure.

Because of ethical concerns, this study did not collect tissue samples for histologic analysis. The current study's findings, however, are in line with recent studies that claimed that PRP cannot significantly influence therapy outcomes on its own. Bezgin et al. [10] came to the conclusion that PRP effectively formed a scaffold for regenerative endodontic treatment, but that there was no appreciable difference in treatment outcomes between PRP and traditional blood clot scaffold. Martin et al., [12] discovered in a different study that PRP may only improve wound healing if parenchymal tissue has not been entirely damaged; otherwise, it may not stimulate tissue regeneration. Because only completely necrotic teeth were included, this would account for the identical results between PRP and the blood clot in the current study.

In a canine study using PRP and dental pulp cells, Zhu et al. [13] discovered that using PRP alone resulted in a decreased rate of dentin wall thickening and less mineralized tissue formation. This outcome was linked to the PRP's growth factors degrading more quickly than expected.

In the previous studies, 3D CBCT has been shown to be more sensitive than periapical radiography in detecting periapical lesion treatment outcomes, it was used in the present study to investigate periapical and root length alterations. To measure radiolucent areas, Estrela et al [11]. successfully used 3D CBCT and periapical index scoring. Bone density was quantitatively assessed in the current study using the software's tools. The size of the lesion was significantly reduced, and bone density increased.

According to Narang et al. [14], the effects of blood clots and PRP on apical closure, root lengthening, dentinal wall thickening, and periapical healing are comparable. In a different investigation, PRP was discovered to be more effective than blood clots at revascularization by Jadhav et al [15]. PRP's effectiveness was credited by the authors to increased collagen formation, sustained growth factor release, and improved periapical mesenchymal and endothelial cell recruitment, retention, and proliferation.

CONCLUSION

Within the limitations of the study, they concluded thatPRP can serve as an effective scaffold for regenerative endodontic therapy. The outcomes of the PRP therapy, however, did not materially differ from those of the standard approach using a blood clot as a scaffold.

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