EVALUATION OF PLATELETS-RICH FIBRIN VERSUS NANOHYDROXYAPATITE AS PULPOTOMY MATERIALS IN PRIMARY MOLARS: A PROSPECTIVE CLINICAL STUDY

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ABSTRACT

Objectives: Pulpotomy is a crucial pulp therapy for severely exposed pulps in primary molars. By preserving the radicular pulp, it tries to keep a healthy tooth in the mouth until its exfoliation. Keeping the tooth healthy and functioning entails removing the coronal pulp and treating the radicular pulp with medication.

Subject and Methods: The children who participated in this clinical investigation ranged in age from 4 to 8 years old and they were chosen from patients undergoing preoperative radiographic and clinical evaluation at the outpatient clinic, forty mandibular primary teeth that required pulpotomy treatment were divided into two groups (n=20) based on the type of pulpotomy material employed, the platelets-rich fibrin group and the Nanohydroxyapatite group. This study was approved as a prospective clinical trial with a split-mouth design. The clinical and radiological outcomes were noted and assessed at two distinct follow-up intervals of three and six months.

Results: The teeth treated with PRF had improved clinical and radiographic outcomes at the two distinct follow-up intervals of 3, and 6 months, but there was no statistically significant difference between them and the teeth treated with NHAp.

Conclusion: The results of the pulpotomized primary molars’ clinical and radiographic outcomes could be improved by using PRF as a pulp dressing agent.

KEYWORDS: Nanohydroxyapatite, platelets-rich fibrin, primary molars, pulpotomy

INTRODUCTION

The coronal pulp becomes inflamed as a result of bacterial invasion if a carious primary molar tooth is left untreated or treated insufficiently (1). The remaining tissue has the potential to recover if the damaged tissue is removed and the radicular pulp stumps are treated with the proper treatment (2). At this point, pulp inflammation is frequently limited to the coronal pulp. When vital pulp therapy is performed to treat severely exposed primary teeth, this facility is used to recover (3). Pulpotomy is a crucial pulp therapy for severely exposed pulps in primary molars. By preserving the radicular pulp, it tries to keep a healthy tooth in the mouth until its exfoliation. Keeping the tooth healthy and functioning entails removing the coronal pulp and treating the radicular pulp with medication (2-4).

New approaches to pulp therapy are now possible thanks to developments in the fields of bone and dentin production. The topical application of platelet concentrates is one of the intriguing pathways (5). The PRF acts as a reservoir for the continual release of growth factor, which controls the process of reparative dentinogenesis, which is the scientific basis for the use of platelet preparation (6). In their study of the impact of PRF on dental pulp

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cells obtained from healthy people having their third molars extracted, Kritik et al (7), found that clinically and radiographically, PRF placement was successful in causing the regeneration of non-vital immature permanent teeth.

An immune concentrate with a certain composition and three-dimensional structure can be thought of as platelet-rich fibrin. It contains a variety of growth factors, including platelet-derived growth factor, transforming growth factor beta 1, and insulin-like growth factor, all of which have a variety of strong local features such as cell migration, adhesion, proliferation, and differentiation (8,9). As PRF is both a healing and an interposition biomaterial, it has been demonstrated to be an appropriate biomaterial for pulp-dentin complex regeneration (5,10).

The regeneration potential of using hydroxyapatite was evaluated as part of our search for the best pulpotomy agent (11). It has been demonstrated that NHAp is a very biocompatible substance for both bone and soft tissues (12). The primary component of tooth hard tissues, hydroxyapatite, has the potential to act as an artificial barrier right away. It has been said to work well for direct pulp capping, osseointegration of titanium implants, repair of periodontal bone abnormalities, and enlargement of the alveolar ridge (13,14).

In our hunt for the ideal pulpotomy agent, we are experimenting with the development of innovative materials that are biocompatible and produce favorable results in the critical pulp treatment procedure known as a pulpotomy. The current study compared platelet-rich fibrin and nanohydroxyapatite crystals as pulpotomy materials in primary molars.

**SUBJECT AND METHODS**

After receiving approval (EC Ref No. 888/3883) from the Faculty of Dental Medicine’s Ethical Committee at Al-Azhar University (Boys, Cairo), this study was carried out. The children aged 4 to 8 years old, who took part in this clinical investigation were chosen from those undergoing preoperative radiographic and clinical evaluations at the outpatient clinic of the Pedodontics and Oral Health Department, Faculty of Dental Medicine, Al- Azhar University (Cairo, Boys) following obtaining signed informed consent from their parents.

Based on a previous study, by Vivek et al (15), the sample size was calculated to be 40 teeth (20 for each group) with a 0.4 effect size, and power of 80%. In 80% (the power) of those experiments, the P value will be less than 0.05 (two-tailed) so the results will be deemed “statistically significant”. In the remaining 20% of the experiments, the increase in survival proportion will be deemed “not statistically significant”.

A total of 40 mandibular primary molar teeth with advanced caries and pulpotomy indications participated in this investigation. Patients who required pulpotomy treatment had at least two primary mandibular molars, one on each side, with essential pulp exposure from caries or mechanical trauma.

After a preoperative radiographic and clinical evaluation, patients were chosen based on the following inclusion and exclusion criteria: symptomless teeth with deep carious lesions, a tooth with a control time of bleeding of no more than five minutes, and the involved teeth should not have any radiographic or clinical signs of pulp degeneration (16,17).

Based on the type of pulpotomy material used, the PRF group (Group A) and the NHAp (NanoGate Co., Egypt) group (Group B), 40 mandibular primary teeth that needed pulpotomy treatment were split into two groups (n=20). The split-mouth prospective clinical trial design for this study was authorized. For all of the selected molars in the same patient, the pulpotomy medication was randomly assigned using the envelope draw method (18). Two distinct follow-up intervals of three, and six months were used to note and evaluate the clinical and radiological outcomes.
Preparation of PRF:

The Choukroun et al. (19) technique was used to prepare the PRF. Without using an anticoagulant, a 1 ml sample of blood was taken from the child’s forearm and transferred to a test tube. The blood sample was immediately centrifuged for 10 minutes at 3000 rpm using an 800 D Centrifuge. Following centrifugation, the tube naturally divided into the following three layers: platelet-poor plasma at the top, a fibrin clot with a high platelet content in the middle, and red blood cells at the bottom. The tube was filled with sterile tweezers, which were used to carefully grip and remove the fibrin clot. To release the fluids entrapped in the fibrin matrix and create a highly resilient autologous fibrin membrane, the PRF clot was squeezed between sterile dry gauze. The fibrin membrane was then placed on a sterile glass slide and split in half using a scalpel with a #15 blade and a #3 handle. (Fig. 1)

Restoration Procedures

In group A, the severed pulp stumps were covered with each half of the freshly created PRF membrane using sterile tweezers after hemostasis had been achieved (17). However, in group B, upon the achievement of hemostasis, the NHAp paste was introduced in the pulp chamber In accordance with the manufacturer’s recommendations, NHAp was combined with physiologic saline in a 3:1 ratio. The NHAp paste was then injected into the pulp chamber using a messing gun and crushed over a moist cotton pellet with a condenser. To ensure a thickness of 2 to 3 mm, care was taken to completely seal the pulp tissue with NHAp paste (21) (Fig. 2).

The pulp chamber was then covered with a thick mixture of zinc oxide and eugenol “ZO/E” (DENTSPLY Detrey GmbH, Konstanz, Germany) as an intermediary restorative substance in both groups. The teeth were then ready to receive a suitable stainless-steel crown (SSC) as a final restoration after 24 hours had passed. Glass ionomer cement (GIC) was used to fix crowns after checking their occlusion and adaptability (Medicem, Promedica Dental Material GmbH, Germany).
Follow-up procedures

In a single visit, the clinical procedures were completed step-by-step. In both test groups, the primary molars that were affected underwent clinical and radiographic evaluations throughout the course of the next six months. The same investigator independently carried out each clinical or radiographic assessment. The following indications and symptoms were included on standardized forms, and any of them were considered failures. The clinical success of the implicated pulpotomized primary molar teeth was determined by whether they exhibited none of the following symptoms: no pain on percussion or palpation, no swelling, no fistula formation, and no pathological tooth movement\(^{(16,17)}\).

European recommendations for using dental radiographs on children are followed in the radiographic evaluation\(^{(22)}\). The radiographical success of the implicated pulpotomized primary molar teeth was determined by whether they exhibited none of the following: no enlargement of the periodontal ligament space, no periapical and furcation radiolucency, no pathologic root resorption, and no canal calcification\(^{(16,17)}\).

Statistical analysis

The Chi-square test was utilized to compare all binary outcome data collected at various time points. The correlation between clinical and radiographic outcomes using the kappa coefficient. SPSS statistical version 21 was used for the statistical analysis (Statistics Statistical Procedures Companion, Chicago, IL, USA). A significance level of \(p\) less than 0.05 was used.

RESULTS

The results of the Chi-square (\(X^2\)) test of the two-pulp capping medicaments after restoration for 3 and 6 months showed a non-statistically significant difference in the radiographic success and failure rate among the two studied capping medicaments. (Tables 1 and 2) According to the Kappa coefficient; the number of observed agreements after 3 months: is 72 (90.00\% of the observations). While the number of agreements expected by chance: is 40.0 (50.00\% of the observations). The Kappa agreement index was (0.800) after 3 months, indicating substantial agreement between the two examinations with a confidence interval excluding (0.669 to 0.931). According to the Kappa coefficient; the number of observed agreements after 6 months: is 62 (77.50\% of the observations). While the number of agreements expected by chance: is 40.0 (50.00\% of the observations). The Kappa agreement index was (0.550) after 6 months, indicating moderate agreement between the two examinations with a confidence interval excluding (0.367 to 0.733). (Table 3)

FIG (2) Placement of A) PRF; B) NHAp paste over amputated pulp stumps.
Table (1) Clinical evaluation of the two studied groups after 3 and 6 months:

<table>
<thead>
<tr>
<th>Variable</th>
<th>PRF</th>
<th>NHAp</th>
<th>Chi-square</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 months Success; n (%)</td>
<td>19 (95%)</td>
<td>18 (90%)</td>
<td>0.3604</td>
<td>0.548 ns</td>
</tr>
<tr>
<td>3 months Failure; n (%)</td>
<td>1 (5%)</td>
<td>2 (10%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6 months Success; n (%)</td>
<td>17 (85%)</td>
<td>15 (75%)</td>
<td>0.625</td>
<td>0.429 ns</td>
</tr>
<tr>
<td>6 months Failure; n (%)</td>
<td>3 (15%)</td>
<td>5 (25%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*: significant at p < 0.05. ; non-significant at p >0.05. ns= non-significant.

TABLE (2) Radiographic evaluation of the two studied groups after 3 and 6 months:

<table>
<thead>
<tr>
<th>Variable</th>
<th>PRF</th>
<th>NHAp</th>
<th>Chi-square</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 months Success; n (%)</td>
<td>18 (90%)</td>
<td>17 (85%)</td>
<td>0.0033</td>
<td>0.953 ns</td>
</tr>
<tr>
<td>3 months Failure; n (%)</td>
<td>2 (10%)</td>
<td>3 (15%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6 months Success; n (%)</td>
<td>16 (80%)</td>
<td>14 (70%)</td>
<td>0.533</td>
<td>0.465 ns</td>
</tr>
<tr>
<td>6 months Failure; n (%)</td>
<td>4 (20%)</td>
<td>6 (30%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*: significant at p < 0.05. ; non-significant at p >0.05. ns= non-significant.

TABLE (3) Kappa correlation between clinical and radiographic results after 3 and 6 months

<table>
<thead>
<tr>
<th>Variable</th>
<th>Clinical evaluation</th>
<th>Radiographic evaluation</th>
<th>Kappa index</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 months Success; n (%)</td>
<td>37 (92.5%)</td>
<td>35 (87.5%)</td>
<td>0.800</td>
</tr>
<tr>
<td>3 months Failure; n (%)</td>
<td>3 (7.5%)</td>
<td>5 (12.5%)</td>
<td></td>
</tr>
<tr>
<td>6 months Success; n (%)</td>
<td>32 (80%)</td>
<td>30 (75%)</td>
<td>0.550</td>
</tr>
<tr>
<td>6 months Failure; n (%)</td>
<td>8 (20%)</td>
<td>10 (25%)</td>
<td></td>
</tr>
</tbody>
</table>

DISCUSSION

Inductive pulpotomy and reparative pulpotomy are other names for regenerative pulpotomy. This process promotes radicular pulp healing and the development of a dentine bridge/hard tissue barrier. Additionally, an uninflamed pulp’s odontoclasts could participate in the exfoliative process at the appropriate time and keep it going in a physiologic way. A second-generation platelet concentration is a platelet-rich fibrin. It is purely autologous and speeds up the healing process by assisting in the release of growth factors required for the regeneration of the dentin pulp complex. PRF was chosen as the researched pulp dressing in this investigation instead of NHAp due to its potential to enhance pulp tissue regeneration and differentiation as well as efficient pulp healing. This is a result of the finding that growth factors present in PRF assist tooth morphogenesis, differentiation, and tissue regeneration. In light of the fact that NHAp and PRF fall under this category, they were selected as the two materials to be researched in the current study.
Since hydroxyapatite is a bio-inductive substance utilized in the regeneration of bone deformities, it was chosen as the study’s test substance since it is biocompatible and resembles natural mineral tissues (25). NHAp was recommended for use in this study as a control group due to its alleged promising osteoconductive and dentinogenic potentials (21).

Mandibular primary molars were chosen as the only involved molars in the current study because they are easier to visualize, have less permanent tooth buds overlapping their roots, and have lower primary molars that are more clearly divided than their maxillary counterparts. This allows the researcher to see the radiographic pathology and healing (26). Additionally, the choice of the primary molars was made definitively during surgery based on the amount of time (5 minutes) required for hemostasis following coronal pulp amputation (18). This is so that the physicians’ experience and the veracity of the information provided in the dental history can accurately diagnose the pulp state and predict how the pulpotomy operation will proceed (18).

As the goal of the scoring assessment was to reveal the severity of changes in the involved teeth after the end of treatment, and molars were not counted as “successful” or “failed” based on clinical signs or symptoms only, the involved teeth in this study were monitored both clinically and radiographically in the follow-up periods (18). Additionally, in the current study, radiographic imaging was carried out using periapical radiographs that clearly show both the periapical and furcal areas before the inclusion of the children and during the follow-up because it is the best way to detect root resorption, periapical and periodontal status, and osseous defects that may indicate failure (21). Preoperative periapical radiographs were also employed in this study to examine the periapical area for correct assessment and to avoid follow-up data bias. They served as a baseline for comparisons in various follow-ups (27).

The children chosen to participate in this study ranged in age from 4 to 8 years. This is due to the fact that this age range represents the most advantageous chronological age with long roots, where root resorption has not yet begun or may be low to maintain patient compliance (28).

Because it is the gold standard for isolation and is used to ensure adequate isolation that is necessary to prevent salivary and bacterial contamination and to avoid bias of improper pulp therapy procedure information, the treated mandibular primary molars were isolated with a rubber dam during the pulpotomy procedures in the current study (29).

The PRF clot needs to be used right away after preparation because otherwise it will shrink, become dehydrated, and lose structural integrity. Dehydration alters the biological characteristics of PRF by lowering its growth factor concentration and leukocyte viability (17). Following a set PRF preparation protocol is therefore crucial for the current study.

In the current inquiry, ZOE was picked over the pulp-dressing materials that had been looked at as middle-filling material. This is because, when used as pulp capping material, the commonly used pharmaceutical ZOE paste performs better at long-term disinfection, offers a thermal insulating base, and has sedative and palliative properties than other base materials like GIC (30). Additionally, in order to effectively seal off the affected tooth from microleakage, prevent its fracture, and ensure that the success or failure of the restoration protocol only depended on the tested materials, GIC was chosen in this study to restore the coronal portion of the pulpotomized primary molar teeth, with SSC serving as the final tooth coverage restoration (31).

In this investigation, the clinical and radiographic follow-up was performed at several time points including 3 months and 6 months. This is due to the American Academy of Paediatric Dentistry’s (AAPD’s) recommendation regarding pulp therapy that teeth with pulpotomies undergo clinical and radiological evaluations at least once every three
months (31). Additionally, it was stated that bone deposition in the pretreatment radiolucent areas is proof that the radiographic infectious process of pulpotomized teeth should resolve in six months (32).

After three and six months of follow-up, PRF had marginally greater success rates than NHAp. This finding raises the possibility that PRF could be used as a biological molecule to enhance the regeneration of missing or damaged dental pulp tissues and to induce reparative dentinogenesis (33). These outcomes may also be ascribed to the positive pulpal response, the great potential for healing, and the high regenerative power linked to the usage of PRF in keeping the primary pulp healthy and vital. Moreover, PRF is an autologous biological substance that aids in the release of growth factors required for the regeneration of the dentin pulp complex, speeding up the healing process. These growth factors are in charge of tooth differentiation and morphogenesis, and recapitulation of these processes enables tissue regeneration (17).

Additionally, it has been postulated that the clot that forms at the pulp surface after contact with the PRF clot may also serve as a barrier for irritant compounds that promote internal resorption and inflammation. This could explain why the PRF had a greater success rate (34). This result concurred with those of Erfanparast et al (35), and Abdel Sameia et al (36), who indicated that the ability of a blood clot to effectively stop pulp tissue inflammation is essential for the initial efficacy of crucial pulp treatment.

The high biocompatibility, alkalinity, regenerative power, and superior sealing ability of NHAp were also cited as contributing factors to the increased success rates of primary molars treated with it (27). Additionally, NHAp may function as a chemical cell absorber that encourages osteoblast invasion, according to Haghgoo et al (37). These findings are consistent with those of Adlakha et al (38), who examined the effectiveness of HAp crystals as pulpotomy agents for primary molars and observed that, following a 6-month follow-up period, all molars treated with HAp were clinically successful.

**CONCLUSIONS**

This randomized clinical study’s findings allow for the following conclusion to be made: By using PRF as a pulp dressing agent, pulpotomized primary molars may have better clinical and radiological outcomes. In pulpotomized primary molars, platelet-rich fibrin performed slightly, but not significantly, better clinically and radiographically than NHAp. Primary teeth that have been pulpotomized can benefit from dressing agents that are both PRF and NHAp.

**REFERENCES**


