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RESEARCH ARTICLE

CLINICAL AND RADIOGRAPHIC EVALUATION OF NANOHYDROXYAPATITE AND PLATELETS-RICH FIBRIN AS PULPOTOMY MATERIALS FOR PRIMARY MOLARS

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Abstract

Objectives: Pulpotomy works to retain a healthy tooth in the mouth till exfoliation by protecting the radicular pulp. The coronal pulp must be removed, and the radicular pulp must be given medication, in order to maintain the tooth's health and function.

Subject and Methods: The children who took part in this clinical investigation were chosen aged 4 to 8 years old, following approval from the Ethical Committee and obtaining signed informed consent from their parents. Based on the type of pulpotomy material used, the NHAp group and the PRF group, and 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. Three distinct follow-up intervals of 6 weeks, six, and twelve months were used to note and evaluate the clinical and radiological outcomes.

Results: The teeth treated with PRF had improved clinical and radiographic outcomes at the three distinct follow-up intervals of 6 weeks, 6, and 12 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.

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Introduction:-

If a carious primary molar tooth is ignored or treated improperly, the bacterial invasion will cause the coronal pulp to become inflamed. [(1)] If the injured tissue is removed and the radicular pulp stumps are given the appropriate care, the remaining tissue may heal. [(2)] At this stage, coronal pulp inflammation is typically the only affected pulp. This facility is used to recover when crucial pulp therapy is applied to treat highly exposed primary teeth. [(3)] Pulpotomy is an essential pulp treatment for primary molars with significantly exposed pulps. It works to retain a healthy tooth in the mouth till exfoliation by protecting the radicular pulp. [(4)]

Thanks to advancements in the formation of bone and dentin, new methods of pulp therapy are now possible. One of the intriguing pathways is the topical use of platelet concentrates. [(5)] The PRF serves as a reservoir for growth factor, which regulates the reparative dentinogenesis process and serves as the rationale for the usage of platelet preparation. [(6)]

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One might imagine platelet-rich fibrin as an immunological concentrate with a specific composition and three-dimensional structure. It contains several growth factors, such as platelet-derived growth factor, transforming growth factor beta 1, and insulin-like growth factor, which all have a number of significant local properties such as cell motility, adhesion, proliferation, and differentiation. [7,8] It has been shown that PRF is a suitable biomaterial for pulp-dentin complex regeneration because it functions as both a healing and an interposition biomaterial. [5]

In our quest to find the ideal pulpotomy agent, the regeneration capacity of hydroxyapatite was assessed. [9] Hydroxyapatite has proven to be a remarkably biocompatible material for both bone and soft tissues. [10] Hydroxyapatite, the main building block of dental hard tissues, has the capacity to function as an artificial barrier immediately away. Direct pulp capping, osseointegration of titanium implants, correction of periodontal bone anomalies, and enlargement of the alveolar ridge have all been suggested to benefit from its use. [11,12]

We are experimenting with the creation of novel materials that are biocompatible and deliver positive outcomes in the crucial pulp treatment technique known as a pulpotomy as part of our search for the ultimate pulpotomy agent. In the current study, pulpotomy materials for primary molars—platelet-rich fibrin and nanohydroxyapatite crystals—were compared.

Subject and Methods:-

This study was done following consent (EC Ref No.888/3883) from the Faculty of Dental Medicine's Ethical Committee at Al-Azhar University (Boys, Cairo). After receiving written informed consent from their parents, the boys aged 4 to 8 who participated in this clinical investigation were selected 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).

The sample size was calculated to be 40 teeth (20 for each group), with a 0.4 effect size and an 80% power, based on research conducted by Vivek et al. in 2009 [13]. The results will be regarded as "statistically significant" if the P value is less than 0.05 (two-tailed) in 80% (the power) of those experiments. The increase in survival rate in the remaining 20% of the experiments will be labeled as "not statistically significant.".

This examination involved 40 mandibular primary molar teeth with severe caries and pulpotomy indications. Two or more primary mandibular molars, one on each side, with critical pulp exposure due to caries or mechanical trauma were present in patients who needed pulpotomy treatment.

Patients were preferred based on the following eligibility and exclusion criteria after a preoperative radiographic and clinical evaluation: a tooth with a control time of bleeding of no more than five minutes, a tooth with symptomless deep carious lesions, and the involved teeth should not have any radiographic or clinical signs of pulp degeneration. [14,15]

For the purpose of dividing the 40 mandibular primary teeth that required pulpotomy treatment into two groups based on the pulpotomy material utilized, the NHAp (NanoGate Co., Egypt) group (Group A), and the PRF group (Group B) were used (n = 20). For this study, the split-mouth prospective clinical trial design was approved. Using the envelope draw approach, the pulpotomy medicine was assigned at random to each of the patient's selected molars. [16] The clinical and radiological outcomes were noted and assessed at two separate follow-up intervals of three and six months.

Preparation of PRF:

The PRF was created using the technique described by Choukroun et al. (2001) [17]. A 1 ml sample of blood was drawn from the child's forearm and put into a test tube without the use of an anticoagulant. The blood sample was immediately centrifuged using an 800 D Centrifuge for 10 minutes at 3000 rpm. After centrifugation, the tube spontaneously separated into the top layer of platelet-poor plasma, the middle layer of a fibrin clot with a high platelet content, and the bottom layer of red blood cells. Sterile tweezers were placed into the tube, and they were used to delicately grasp and remove the fibrin clot. The PRF clot was squashed between sterile dry gauze in order to liberate the fluids contained in the fibrin matrix and produce a very resilient autologous fibrin membrane. Then, using a scalpel with a #15 blade and a #3 handle, the fibrin membrane was put on a clean glass slide and cut in two. (**Fig. 1**)

Fig. 1:- Preparation of PRF.

Pulpotomy Procedures

Topical anaesthetic (Xylocaine, AstraZeneca, Sodertalje, Sweden) was applied to all concerned teeth for one minute to start the therapy. After administering the 3% mepivacaine anaesthetic solution (Mepecaine L: Mepecaine Hcl 3%, Alexandria Co. for Pharmaceuticals, Alexandria, Egypt), a nerve block injection was given using aspirating dental syringes and 27-gauge needles. To keep the accused side apart, a rubber dam was deployed. After that, caries was removed while being well-irrigated with a large, slowly rotating round bur. The pulp chamber was then accessed using a carbide fissure bur. A sterile high-speed diamond round bur was utilized to remove the pulp chamber's roof after pulp exposure. The coronal pulp tissue was severed with a sharp spoon excavator, and the pulp chamber was fully irrigated with sterile physiological saline. Moist, sterile cotton pellets were applied to the radicular pulp's entrance and held there for 5 minutes to establish stasis. If the bleeding did not stop after five minutes, the tooth was pulped and taken out of the study. [15,17]

Restoration Procedures

After hemostasis was achieved in group A, the NHAp was mixed in a 3:1 ratio with physiologic saline in accordance with the manufacturer's instructions. A messing gun was used to inject the NHAp paste into the pulp chamber, and a condenser was used to crush the paste over a damp cotton pellet. Care was made to thoroughly seal the pulp tissue with NHAp paste in order to provide a thickness of 2 to 3 mm. [18] In contrast after hemostasis was accomplished in group B, the severed pulp stumps were covered with each half of the newly made PRF membrane using sterile tweezers. [15] (**Fig. 2**)

As an intermediate restorative ingredient in both groups, the pulp chamber was then covered with a thick mixture of zinc oxide and eugenol (ZOE) (DENTSUPLY Detrey GmbH, Konstanz, Germany). After 24 hours, the teeth were prepared for an appropriate stainless-steel crown (SSC) as a final restoration. Crowns were fixed using glass ionomer cement (GIC) after being examined for occlusion and adaptability (Medicem, Promedica Dental Material GmbH, Germany).

Fig. 2:- Laying of A); NHAp paste B) PRF over remnants of pulp from amputation.

Follow-up procedures

The clinical procedures were carried out sequentially in a single visit. Over the following six months, the primary molars in both test groups that were impacted received clinical and radiographic assessments. Each clinical or radiographic evaluation was performed independently by the same investigator. Any of the following signs and symptoms were listed on standardized forms and were regarded as failures. If the implicated pulpotomized primary molar teeth showed any of the following symptoms—pain on percussion or palpation, edoema, the development of fistulas, or pathological tooth movement—clinical success was assessed. [14,15]

The radiographic evaluation complies with European criteria for the use of dental radiographs on children. [19] The radiographic success of the involved pulpotomized primary molar teeth was assessed by looking for none of the following signs: canal calcification, periapical and furcation radiolucency, widening of the periodontal ligament space, and periapical and furcation radiolucency. [14,15]

Statistical analysis:

All binary outcome data gathered at various times were compared using the Chi-square test. using the kappa coefficient to determine the relationship between clinical and radiographic results. The statistical analysis was performed using SPSS statistics version 21 (Statistics Statistical Procedures Companion, Chicago, IL, USA). The significance level was set at p less than 0.05.

Results:-

The results of the Chi-square test of the two-pulp capping medicaments after restoration for 6 weeks, 6 and 12 months showed a non-statistically significant difference in the radiographic success and failure rate among the two studied capping medicaments. (**Tables 1 and 2**) After six weeks, the Kappa agreement index showed nearly perfect agreement between the two tests with a confidence range excluding (1.000 to 1.000). While after six months, the Kappa agreement index showed moderate agreement between the two exams with a confidence interval excluding (0.367 to 0.733). However, after 12 months, the Kappa agreement index showed fair agreement between the two exams with a confidence interval excluding (0.146 to 0.554) (**Table 3**)

Table (1):- Clinical evaluation of the two studied groups after 6 weeks, and 6, and 12 months:

Variable		NHAp	PRF	Chi-square	p-value
6 weeks	Success; n (%)	20 (100%)	20 (100%)	0.0	1 ns
	Failure; n (%)	0 (0%)	0 (0%)		
6 months	Success; n (%)	15 (75%)	17 (85%)	0.625	0.429 ns
	Failure; n (%)	5 (25%)	3 (15%)		
12 months	Success; n (%)	13 (65%)	16 (75%)	0.476	0.490 ns
	Failure; n (%)	7 (35%)	4 (25%)		

*; significant at $p < 0.05$.

; non-significant at $p > 0.05$. ns= non-significant.

Table (2):- Radiographic evaluation of the two studied groups after 6 weeks, and 6, and 12 months:

Variable		NHAp	PRF	Chi-square	p-value
6 weeks	Success; n (%)	20 (100%)	20 (100%)	0.0	1 ns

	Failure; n (%)	0 (0%)	0 (0%)		
6 months	Success; n (%)	14 (70%)	16 (80%)	0.533	0.465 ns
	Failure; n (%)	6 (30%)	4 (20%)		
12 months	Success; n (%)	11 (55%)	14 (70%)	0.96	0.327 ns
	Failure; n (%)	9 (45%)	6 (30%)		

*; 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:

Variable		Clinical evaluation	Radiographic evaluation	Kappa index
6 weeks	Success; n (%)	40 (100%)	40 (100%)	1.000
	Failure; n (%)	0 (0%)	0 (0%)	
6 months	Success; n (%)	32 (80%)	30 (75%)	0.550
	Failure; n (%)	8 (20%)	10 (25%)	
12 months	Success; n (%)	29 (72.5%)	25 (62.5%)	0.350
	Failure; n (%)	11 (27.5%)	15 (37.5%)	

Discussion:-

Due to its capacity to promote pulp tissue regeneration and differentiation as well as effective pulp healing, PRF was chosen as the investigated pulp dressing in this study instead of NHAp. The discovery that growth factors contained in PRF support tooth morphogenesis, differentiation, and tissue regeneration led to this conclusion. [20] NHAp and PRF were chosen as the two materials to be examined in the current study since they fall into this group.

Moreover, it was decided to use hydroxyapatite as the study's test substance since it is biocompatible and resembles natural mineral tissues, and is a bio-inductive substance used in the regeneration of bone abnormalities. [21] The choice of NHAp as a control group in this investigation was advised because of its purportedly promising osteoconductive and dentinogenic potentials. [19]

The age range of the kids selected to take part in this study was 4 to 8. This is because root resorption has not yet started or may be low at this age range, which is the most favourable chronological age with lengthy roots. [22]

Because they are simpler to see, have fewer permanent tooth buds overlapping their roots, and have lower primary molars that are more clearly split than their maxillary counterparts, mandibular primary molars were chosen as the only involved molars in the current investigation. This enables the researcher to see the disease and healing shown on radiographs. [23] In addition, the decision of the primary molars was established to be included based on how long it took for hemostasis to occur after coronal pulp amputation. [16]

The involved teeth in this study were monitored both clinically and radiographically during the follow-up periods as the scoring assessment's objective was to reveal the severity of changes in the involved teeth after the end of treatment. Molars were not classified as "successful" or "failed" based solely on clinical signs or symptoms. [16] Furthermore, radiographic imaging was utilized in the current study using periapical radiographs that clearly depict 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. [19] This study also used preoperative periapical radiographs to investigate the periapical region for accurate assessment and to prevent follow-up data bias. They served as a reference point for comparisons in different follow-ups. [24] The clinical and radiological follow-up in this study was carried out at several intervals, including 6 months. This is because it was suggested that the radiographic infectious process of pulpotomized teeth should clear in six months due to bone deposition in the pretreatment radiolucent areas. [25]

ZOE was chosen for the investigation as middle-filling material because the frequently used medicinal ZOE paste, when used as pulp capping material, outperforms other base materials like GIC in terms of long-term disinfection, provides a thermal insulating base, and has sedative and palliative qualities. [26] Additionally, GIC was chosen in this study to restore the coronal portion of the pulpotomized primary molar teeth, with SSC acting as the final tooth coverage restoration, in order to effectively seal off the affected tooth from microleakage, prevent its fracture, and guarantee that the success or failure of the restoration protocol only depended on the tested materials. [27]

The positive results of the NHAp and PRF groups after 1.5 months of observation are consistent with a study by Mostafaa et al. (2018) [15] which revealed the absence of pain and mobility, as all teeth were asymptomatic after 3 months (not just 1.5 months) of observation and PRF demonstrated a 100% success rate. Also, Adlakha et al. (2009) [28] examined the effectiveness of HAp crystals as pulpotomy agents for primary molars and found that, after a 6-month follow-up period, all molars treated with HAp were clinically successful. This is due to the well-established function played by bacterial contamination in preventing pulp healing following essential pulp therapy. [29]

Following six and twelve months, PRF had somewhat higher success rates than NHAp. The prospect that PRF could be utilized as a biological molecule to promote the regeneration of missing or injured dental pulp tissues and to trigger reparative dentinogenesis is suggested by this finding. [30] These results can also be attributed to the PRF's use in maintaining the primary pulp's health and vitality, which has a favourable pulpal response, significant healing capacity, and high regenerative power. Furthermore, PRF, an autologous biological component, helps release growth factors needed for the regeneration of the dentin pulp complex, hastening the healing process. The differentiation and morphogenesis of teeth are controlled by these growth factors, and recapitulation of these processes promotes tissue regeneration. [15] The clot that forms at the pulp surface following contact with the PRF clot may also operate as a barrier for irritating substances that encourage interior resorption and inflammation. [31] This may be the reason the PRF had a higher success rate.

The higher success rate of primary molars treated with NHAp was attributed to their high biocompatibility, alkalinity, regeneration ability, and superior sealing ability. [23] These results support those of Adlakha et al. (2009) [28] who investigated the efficiency of HAp crystals as pulpotomy agents for primary molars and found that, after a 6-month follow-up period, all molars treated with HAp were clinically successful.

Conclusions:-

The results of this randomized clinical investigation allow us to draw the following conclusion: Pulpotomized primary molars may achieve improved clinical and radiological results by employing PRF as a pulp dressing agent. Platelet-rich fibrin performed marginally, but not substantially, better clinically and radiographically than NHAp in pulpotomized primary molars. dressing agents that are both PRF and NHAp can assist primary teeth that have had pulpotomy.

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