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Healing Assessment of Osseous Defects after Surgical Removal of Periapical Lesions in the Presence of Hydroxyapatite, Nanohydroxyapatite, and a Combination of Nanohydroxyapatite and Platelet-rich Fibrin: A Clinical Study

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Abstract

AIM: This study aims to evaluate the bone healing in failed endodontically treated teeth after surgical removal of periapical lesions and placement of hydroxyapatite (HA), nanohydroxyapatite (nHA), and a combination of nHA with platelet-rich fibrin (PRF) periapically.

METHODS: The study was conducted on 24 patients having periapical radiolucency in single-rooted teeth. The selected teeth were divided into three groups: Group A, Group B, and Group C; of eight teeth each. All the teeth were retreated in two visits. In the first visit, the old filling was removed using ProTaper retreatment files (Dentsply Sirona®) then irrigation with sodium hypochlorite 2.5% was done. All canals were dried and filled with di-antibiotic paste (metronidazole and ciprofloxacin). In the second visit, the canals were obturated with ProTaper gutta-percha points and root canal sealer (ADSEAL resin sealer) followed by surgical intervention in the same day. A periapical curettage along with apicoectomy was established. In all the groups, root end cavity was prepared and filled with MTA (ProRoot MTA; DENTSPLY Tulsa Dental Specialties). In Group A, HA powder was packed in the curetted periapical defect. In Group B, nHA powder was packed in the curetted periapical defect. In Group C, nHA with PRF was mixed and packed in the curetted periapical defect. In all groups, patients recall visits were scheduled at 1, 3, and 6 months' time intervals for clinical and radiological evaluation.

RESULTS: After 1 month, there was a statistically significant difference between the median percentage changes in lesions size in the three groups. Pairwise comparisons between groups revealed that there was no statistically significant difference between Group B (nHA) and Group C (PRF and nHA). Both showed statistically significantly higher median percentage reduction in lesions size than Group A (HA group). After 3 as well as 6 months; there was no statistically significant difference between the median percentage decreases in lesions size in the three groups.

CONCLUSION: It was concluded that nHA combination with PRF produced faster periapical healing (bone regeneration) in the first 3 months than nHA alone. However, HA produces periapical healing (bone regeneration) after 6 months.

Introduction

Bacterial infection of the dental pulp may lead to periapical lesions [1]. Most periapical lesions (>90%) can be classified as dental granulomas, radicular cysts, or abscesses [2], [3]. The success of endodontic therapy depends on complete periapical repair and regeneration.

Most of the time teeth with periapical lesions heal satisfactorily after non-surgical endodontic intervention [4]. However, we do come across cases with persisting symptoms and infection that require periradicular surgery to remove the pathological tissues, eliminate the source of irritation, and promote healing [5]. Bone regeneration is a very slow process. To induce bone regeneration and soft tissues healing after oral surgery, the local application of hormones, growth factors, and plasma derivatives has been advocated.

Platelet-rich fibrin (PRF) has been successfully used with bone grafts for bone regeneration in the treatment of periapical defects [6].This autologous matrix is a rich source of growth factors and its application is thought to be an effective way of inducing tissue repair and regeneration. It has the ability to stimulate a cascade of healing events, which can result in the excellent osseous defect fill on radiographic evaluation [6].

Many materials were used as bone grafts to enhance bony healing such as bioactive calcium phosphate ceramics which are the largest family of alloplastic materials, that is, hydroxyapatite (HA) and tricalcium phosphate. They had been used several times for enhancement of bone fill after periapical surgery [7], [8], [9], [10]. However, many researchers have

suggested the use of bone substitutes to fill the space created after removal of periapical pathological lesions surgically to speed up the healing process [11], [12].

Recently, nanotechnology was introduced in dentistry for the creation and utilization of materials, devices, and systems through the control of matter on the nanometer-length scale, that is, at the level of atoms, molecules, and supramolecular structures to improve the material properties [13].

Nanohydroxyapatite (nHA) has unique properties related to its small size and large surface area. That's why we used nHA and its combination with PRF in the present study for bone regeneration in periapical area. The null hypothesis that there is no difference would be found in bone healing between HA, nHA, and its combination with PRF groups.

Subjects and Methods

Sample size calculation

A power analysis was designed to have adequate power to apply a statistical test of the null hypothesis that there is no difference would be found in bone healing between different groups. By adopting an alpha level of 0.05 a beta of 0.2, that is, power=80% and an effect size (f) of 0.64 calculated based on the results of Johns *et al*. [14]; the predicted sample size (n) was a total of 24 samples. Sample size calculation was performed using G*Power version 3.1.9.7 [15].

Materials and devices

1. HA powder (J.T. Baker chemical company)
2. Nano-HA powder (NanoTech Egypt for Photo-Electronics, Giza, Egypt)
3. Centrifuge device (Jiangsu Jinyi Instrument Technology Co., China).

Inclusion criteria for patients according to which patients were selected for the study

- Should be willing to cooperate in this study and ready for presenting regularly for follow-up visits up to the end of the study period
- Should be free from any systemic chronic debilitating diseases that may contraindicate the use of local anesthesia, or has negative effect on healing such as uncontrolled diabetes, immune diseases, uncontrolled cardiovascular diseases, and blood coagulation problems
- Patients suffering from a necrotic tooth with periapical lesions starting from 5 mm or more in diameter related to failed endodontically treated single canalled teeth
- Patients were informed in details about the procedure and the materials used and were asked to sign a written consent form
- All reasonable steps to protect the security of personal information and privacy of the patient protected health information were taken
- All data were kept confidential and the faculty of research ethics committee reviewed the proposal.

Exclusion criteria

- Patients taking chemotherapy or radiotherapy to the head-and-neck region in the past 12-month period
- Patients taking any drugs which may affect the healing process, for example, systemic steroids or anticoagulant therapy
- Patients with bad oral hygiene and periodontal problems.

The present *in vivo* study was conducted on 24 patients having periapical radiolucency related to single-rooted teeth which were selected according to inclusion and exclusion criteria from the outpatient clinic of Endodontic Department of Faculty of Dental Medicine, Minia University. Ethical clearance was taken from the ethical committee of the Institute with approval number: 238.

Periapical radiographs and CBCT were taken for all patients to determine the approximate size of the intra bony defect (the estimated size was measured from cross-sectional, sagittal, and coronal views) and it was approximate, because the measurements were taken from three different dimensions, views, or angulations and the mean of the three measurements gave us the defect's size.

All patients were suffering from a necrotic tooth with periapical lesions starting from 5 mm or more in diameter related to failed endodontically treated single canalled teeth with healthy periodontal status.

Patients with total of 24 affected single-rooted teeth were randomly divided into three groups with a total of eight teeth each.

Procedure

First visit

Periapical radiographs were taken for all the patients. All patients were anesthetized by buccal infiltration technique using local anesthesia; articaine in 4% solution with epinephrine in concentration of 1:100,000. An access cavity in the lingual surface of the anterior teeth was prepared. Rubber dam was placed. Working length was determined by periapical radiographs and an apex locator (Woodpecker® Europe - Poland - Wroclaw). Cleaning and shaping were

done in a crown-down technique using ProTaper rotary files mounted on E-Connect endomotor (Eighteeth® Changzhou, China).

Root canals were irrigated with sodium hypochlorite (2.5% concentration) in stable cases, while in swelling cases, first with saline and then followed with sodium hypochlorite (2.5% concentration) later on in other visits. Then, root canals were dried and filled with di-antibiotic paste (metronidazole and ciprofloxacin) using lentulo spiral. A piece of cotton was placed in the pulp chamber and the access was sealed with resin-modified glass ionomer for 10 days.

Second visit

Patients were examined clinically after 10 days to ensure that there was no swelling, exudate, or foul odor with paper points or pain with percussion. If there were no symptoms, the patients were instructed to rinse with 0.2% chlorhexidine mouth wash 1 day before the day of obturation and surgery. If the patient complained of symptoms, canals were irrigated with sodium-hypochlorite, dried, filled with di-antibiotic paste for 10 days, a piece of cotton was placed in the pulp chamber and the access sealed. Usually, this protocol continued for a period of time depending on the virulence factor of the bacteria and patient's immunity to give a chance for the intracanal medication to produce its effect. The majority of cases took from 4 to 6 weeks for the symptoms to subside. Finally, the canals were obturated with ProTaper gutta-percha points and root canal sealer (ADSEAL resin sealer) followed by the surgery in the same day.

Periapical surgery was performed under strict aseptic conditions. Profound buccal and palatal/lingual infiltration with local anesthesia was injected using articaine 4% solution with 1:100,000 epinephrine. After 15 min, a full mucoperiosteal flap (modified rectangular flap) was raised. Apical curettage and root end resection with fissure bur mounted to 45° surgical handpiece with saline irrigation were performed. Finally, root end cavity was prepared with ultrasonic tips and filled with MTA.

Group A: HA powder was packed in the bony cavity using. Group B: nHA powder was packed in the bony cavity using condenser. Group C: The PRF gel was prepared by the protocol set by Choukroun *et al*. [16]. A small amount of the patient's own blood (10–40 ml) was collected, before the surgical operation, into sterile-dried Monovettes without an anticoagulant. The collected blood was immediately centrifuged for 10 min at 2500 rpm. It was then settled into the following layers: Red lower fraction containing red blood cells, upper straw-colored cellular plasma, and the middle fraction containing the fibrin clot. The resulting clot was extracted from the container using thin sterile forceps and entirely placed in a sterile glass container (Figure 1). Then the nano-HA powder was mixed with

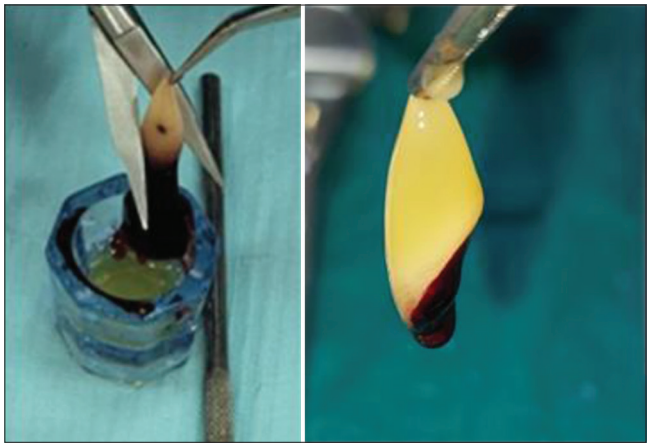


Figure 1: Platelet-rich fibrin obtained from patient's blood

previously prepared PRF and packed in the bony cavity using condenser.

Wound was sutured, patients were instructed to apply cold compresses 15 min every 1 h for 3 hours (at the 1st day of surgery). A course of antibiotics (augmentin 1 g every 12 h and Flagyl 500 mg every 8 h for 5 days), together with anti-inflammatory and analgesic (Bi-Profenid 150 mg every 8 h for 5 days) was given orally. Patients were instructed to rinse their mouth 3 times daily (1 day after surgery) with warm saline and 0.1% chlorhexidine gluconate for 10 days. All sutures were removed after 10 days.

In all groups, patients recall visits were scheduled after 1, 3, and 6 months for clinical and radiological examination (Tables 1-3). On each visit, the patient was examined clinically regarding postoperative discomfort, pain, sensitivity to percussion, and presence/absence of swelling. Digital radiographs were obtained throughout the study for all groups (A, B, and C) with a paralleling technique [17]. Using size 2 charged couple device intraoral digital sensor with XCP, RINN crop film holding system. The sensor was positioned

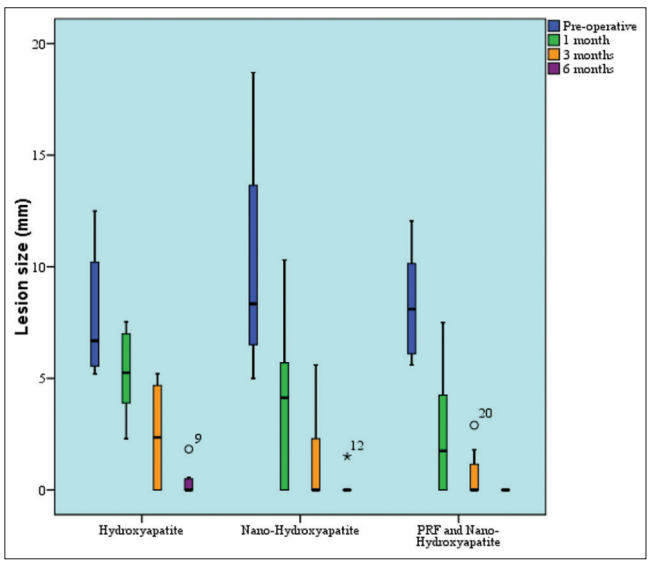


Figure 2: Box plot representing median and range values for lesions size in different groups (circles and star represent outliers)

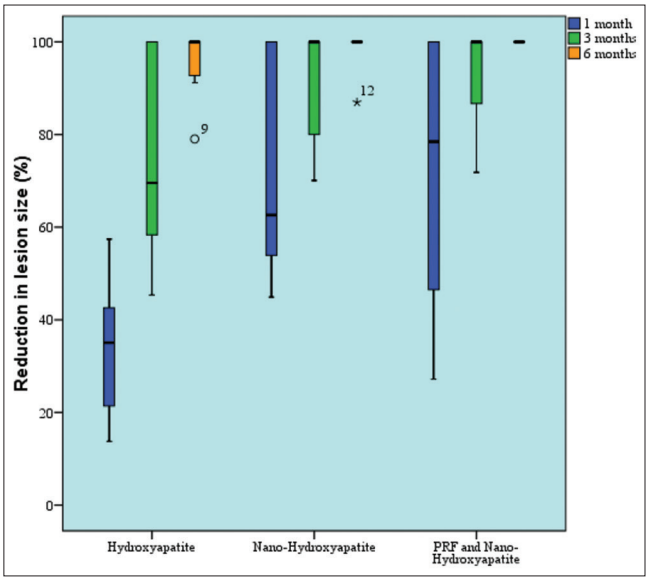


Figure 3: Box plot representing median and range values for percentage reduction in lesions size (%) in the three groups (star and circle represent outliers)

in the mouth parallel to the long axis of the tooth being imaged. The X-ray tube was aimed at the right angle to both the tooth and the sensor. After 6 months, CBCT was taken to evaluate bone density and confirm bone healing. Linear radiographic measurements (in mm) were made on digital periapical radiographs to assess the surface area of the bony defect and compared with pre-operative radiograph [18], [19], [20].

Numerical data were explored for normality by checking the distribution of data and using tests of normality (Kolmogorov–Smirnov and Shapiro–Wilk tests). Data showed non-normal (non-parametric) distribution. Data were presented as mean, standard deviation, median, and range values. Kruskal–Wallis test was used to compare between the three groups. Friedman's test was used to study the changes by time within each group. Dunn's test was used for pairwise comparisons when Kruskal–Wallis or Friedman's test is significant. The significance level was set at $p \leq 0.05$. Statistical analysis was performed with IBM SPSS Statistics for Windows, Version 23.0. Armonk, NY: IBM Corp.

Results

Lesions size (mm)

Comparison between the groups

Preoperatively, after 1, 3, as well as 6 months; there was no statistically significant difference between median lesion sizes in the three groups ($p = 0.554$, effect size = 0.079), ($p = 0.122$, effect size = 0.053), ($p = 0.205$, effect size = 0.007), and ($p = 0.096$, effect size = 0.073), respectively (Table 4).

Changes within each group

In HA as well as nano-HA groups; there was a statistically significant change in lesion size by time ($p < 0.001$, effect size = 0.978) and ($p < 0.001$, effect size = 0.878), respectively. Pairwise comparisons revealed that there was a statistically significant decrease in lesion size after 1 month, from 1 to 3 as well as 3 to 6 months (Figure 2).

In PRF and nano-HA group; there was a statistically significant change in lesion size by time ($p < 0.001$, effect size = 0.875). Pairwise comparisons revealed that there was a statistically significant decrease in lesion size after 1 month as well as from 1 to 3 months. From three to 6 months; there was no statistically significant change in lesion size (Table 5).

Percentage reduction in lesions size

Percentage reduction in lesions size was calculated as follows:

[(pre-operative size-post-operative size)/pre-operative size × 100]

After 1 month; there was a statistically significant difference between median percentage changes in lesions size in the three groups ($p = 0.003$, effect size = 0.382). Pairwise comparisons between groups revealed that there was no statistically significant difference between nano-HA and PRF and nano-HA groups; both showed statistically significantly higher median percentage reduction in lesions size than HA group.

After 3 as well as 6 months; there was no statistically significant difference between median percentage decreases in lesion sizes in the three groups ($p = 0.077$, effect size = 0.092) and ($p = 0.096$, effect size = 0.073), respectively (Figure 3 and Table 6).

Discussion

The ultimate goal of periapical surgery is the predictable regeneration of periapical tissues, including complete repair of osseous defects. Inadequate bone healing is caused by ingrowth of connective tissue into the bone space, preventing osteogenesis. To prevent this soft-tissue ingrowth, bone grafts can be used to fill the bony space in case of large bony defects [21]. Ideal wound healing would achieve maximum regeneration and minimal repair so that the biological function of the injured tissue would not be jeopardized.

Healing of the decayed tissue either by regeneration or by repair depends on the availability of cell types needed and the presence or absence of signals necessary to stimulate these cells. Different

Table 1: Group A case radiographs

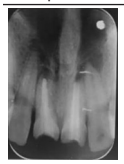
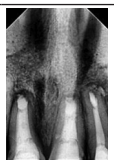
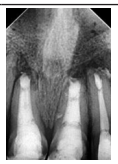
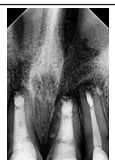
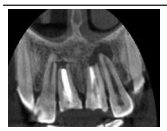
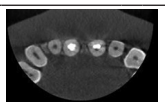
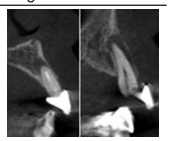
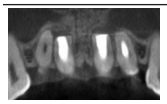
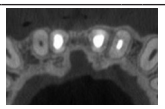
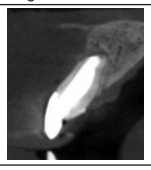
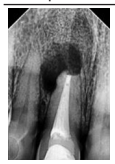
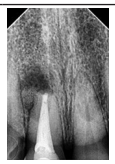
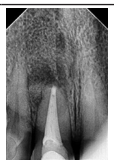
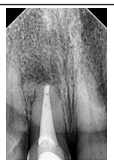
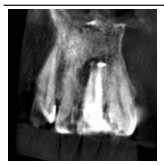
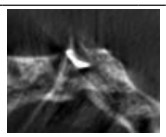
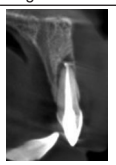
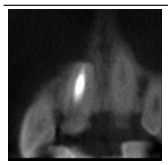
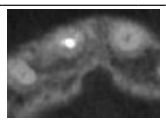
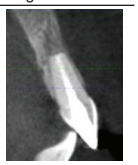
Pre-operative	After 1 month	After 3 months	After 6 months
			
CBCT Before			
Coronal	Axial		Sagittal
			
After			
Coronal	Axial		Sagittal
			

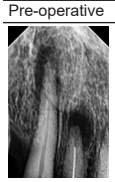
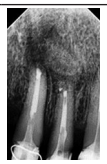
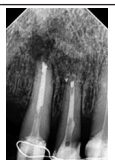
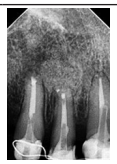
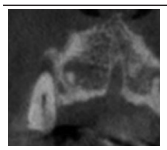
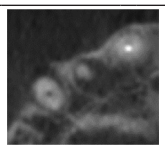
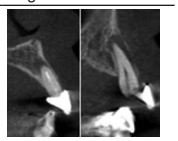
Table 3*:* Group C case radiographs

Pre-operative	After 1 month	After 3 months	After 6 months
			
CBCT Before			
Coronal	Axial		Sagittal
			
After			
Coronal	Axial		Sagittal
			

bone grafts with different properties have been studied and tried in various cases including bioactive calcium phosphates as HA [22].

In this study, we compared the healing efficacy of bone when used HA, nHA, and combination of nHA with PRF as bone graft after the removal of periapical lesion. HA is one of the most widely used alloplastic grafts in both the research and clinical fields. It has a similar composition and structure to natural bone mineral and is known to chemically bond directly to bone when implanted [23]. The properties of HA ceramics can be improved by controlling important parameters of powder precursors such as particle size, particle distribution, and agglomeration [24]. The smaller the particle size, the larger the surface area in volume. Because the cells are too big for the small pores, blood plasma containing all the important proteins is

Table 2*:* Group B case radiographs

Pre-operative	After 1 month	After 3 months	After 6 months
			
CBCT Before			
Coronal	Axial		Sagittal
			
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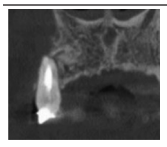	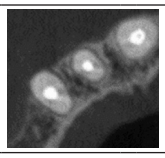		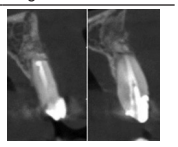

retained in the interstices. The surface of the pores and also of the nanopores is modified in such a way that it “hangs on” to the proteins. Features of nano-bone graft materials include osteoinductive, fully synthetic, highly porous, nanostructured, absorbs natural proteins into the nanopores, should be degraded by osteoclasts, and should have good processability [25], [25], [26], [27].

Nanohydroxyapatite (nHA) possesses unique properties related to its small size and large surface area [28]. Biomaterial researches have focused on the use of nanotechnology to enhance bioreactivity. Thus, bone grafts of synthetic nHA crystals are widely utilized in the repair of bony defects [29].

Favorable osteogenic capacity and enhancement of bone regeneration of nHA were explained as the nanosized particles and their structural similarity to natural bone allows HA nanocrystals to bond firmly to bone. This stimulates the proliferation and metabolism of osteoblasts permitting better osseointegration and strong osteoconductive and osteoinductive ability [30]. It was suggested that when nHA starts to dissolve, it releases calcium ions in the surrounding environment which encourages cell proliferation and osteogenic differentiation. Researchers added that nHA can augment mineral deposition promoting rapid neo-osteogenesis [31]. It was speculated that if HA was placed underneath healthy periosteum and properly vascularized bone, it will release phosphate ions into the medium inducing bone healing [32]. Investigators proposed that nHA induces alveolar osteoblasts to secrete specific bone morphogenic proteins and other growth factors which stimulate and regulate bone regeneration process [32].

Choukroun’s PRF, a second-generation platelet concentrate, is an autologous leukocyte and PRF material. This is produced in a natural manner,

Table 4: Descriptive statistics and results of Kruskal–Wallis test for comparison between lesions size (mm) in the three groups

Time	Hydroxyapatite		Nanohydroxyapatite		PRF and nanohydroxyapatite		p-value	Effect size
	Mean (SD)	Median (range)	Mean (SD)	Median (range)	Mean (SD)	Median (range)		(Eta squared)
Pre-operative	7.9 (2.8)	6.7 (5.2–12.5)	10.2 (4.8)	8.3 (5–18.7)	8.3 (2.4)	8.1 (5.6–12.1)	0.554	0.079
1 month	5.2 (1.9)	5.3 (2.3–7.5)	3.5 (3.7)	4.1 (0–10.3)	2.4 (2.9)	1.8 (0–7.5)	0.122	0.053
3 months	2.3 (2)	2.4 (0–5.2)	1.3 (1.9)	0 (0–5.6)	0.7 (1.1)	0 (0–2.9)	0.205	0.007
6 months	0.3 (0.6)	0 (0–1.8)	0.2 (0.5)	0 (0–1.5)	0 (0)	0 (0–0)	0.096	0.073

*Significant at P ≤ 0.05, PRF: Platelet-rich fibrin.

without adding anticoagulant neither bovine thrombin nor calcium chloride during blood harvest. The absence of anticoagulant implies the activation in a few minutes of most platelets of the blood sample in contact with the tube walls and release of the coagulation cascades. The protocol is very simple and of low cost [33]. PRF is a matrix of autologous fibrin, in which large quantity of platelet and cytokines is embedded intrinsically leading to their progressive release over time, as the network of fibrin disintegrates [34], [35], [36], [37].

In this study, PRF was used in the form of a platelet gel which offers several advantages including promoting wound healing, bone growth and maturation, graft stabilization, wound sealing, hemostasis, and improving the handling properties of graft materials [6]. PRF is a concentrated suspension of the growth factors found in platelets. These growth factors are involved in wound healing and are postulated as promoters of tissue regeneration. PRF is a rich source of platelet-contained growth factor (PDGF), transforming growth factor (TGF), and insulin-like growth factor (IGF). IGF-I stimulates bone formation by proliferation and differentiation, and it is synthesized and secreted by osteoblasts [38]. An increase in the proliferation of human osteoblasts has been demonstrated with a combination of PDGF, IGF-I, TGF, and epidermal growth factor [39].

In the present study, combination of nHA and PRF is used because it helps in faster bone regeneration and prevents scar tissue formation. As supported by recent study that evaluated bone regeneration in the periapical region using platelet-rich fibrin (PRF) and nanocrystalline HA with collagen in combination with PRF and their effects on healing and concluded that the combination of PRF and nanocrystalline HA with collagen produced a significantly faster bone regeneration and that conventional technique and PRF were less predictable with its healing response and bone regeneration [40], [41].

In the present study, many other common factors in all groups aided in success of surgical treatment. Apical seal can be obtained by the use of root-end filling materials [42]. MTA is preferred as retrograde filling material over other materials. MTA has shown the highest healing rates (91.4%) in comparison to other root-end filling materials [43]. MTA also shows less leakage than other root-end filling materials [44].

The crown-down technique was used for cleaning and shaping because it permits straight access to the apical region, eliminates coronal interferences, removes the bulk of tissue and microorganisms before apical shaping, allows deeper penetration of irrigants, and allows better control over working length [45].

NaOCl was used as an irrigant because of its broad-spectrum antimicrobial activity as well as its capacity to dissolve necrotic tissue remnants [46].

The double antibiotic paste was used as intra canal medication between visits. Local application of antibiotics has been investigated as intracanal medicaments [47]. The triple antibiotic paste is a combination of metronidazole, ciprofloxacin, and minocycline. It was shown to be effective in reducing viable bacteria in regenerative protocols [48]. In double antibiotic paste, minocycline is not included to overcome the discoloration caused by the triple antibiotic paste and both triple and double antibiotic pastes have antibacterial effect on human dentine [49].

In the present study, it was found that HA as well as nano-HA groups; there was a statistically significant change in lesions size by time. Pairwise comparisons revealed that there was a statistically significant decrease in lesions size after 1 month, from 1 to 3 as well as 3 to 6 months.

It was found that in combination of PRF with nano-HA group; there was a statistically significant change in lesion sizes by time. Pairwise comparisons revealed that there was a statistically significant decrease in lesions size after 1 month as well as from 1 to 3 months. From 3 to 6 months; there was no statistically significant change in lesions size.

Comparison between the groups revealed that there was no statistically significant difference between combination of nano-HA with PRF group and nano-HA groups after 1 month; both showed statistically

Table 5: Descriptive statistics and results of Friedman's test for comparison between lesions size (mm) at different follow-up times within each group

Time	Hydroxyapatite		Nanohydroxyapatite		PRF and nanohydroxyapatite	
	Mean (SD)	Median (range)	Mean (SD)	Median (range)	Mean (SD)	Median (range)
Pre-operative	7.9 (2.8)^{A}	6.7 (5.2–12.5)	10.2 (4.8)^{A}	8.3 (5–18.7)	8.3 (2.4)^{A}	8.1 (5.6–12.1)
1 month	5.2 (1.9)^{B}	5.3 (2.3–7.5)	3.5 (3.7)^{B}	4.1 (0–10.3)	2.4 (2.9)^{B}	1.8 (0–7.5)
3 months	2.3 (2)^{C}	2.4 (0–5.2)	1.3 (1.9)^{C}	0 (0–5.6)	0.7 (1.1)^{C}	0 (0–2.9)
6 months	0.3 (0.6)^{D}	0 (0–1.8)	0.2 (0.5)^{D}	0 (0–1.5)	0 (0)^{C}	0 (0–0)
p-value	<0.001*		<0.001*		<0.001*	
Effect size (w)	0.978		0.878		0.875	

*Significant at P ≤ 0.05, different superscripts in the same column indicate statistically significant change by time, SD: Standard deviation, PRF: Platelet-rich fibrin.

Table 6: Descriptive statistics and results of Kruskal–Wallis test for comparison between percentage reduction in lesions size (%) in the three groups

Time	Hydroxyapatite		Nanohydroxyapatite		PRF and nanohydroxyapatite		p-value	Effect size (Eta squared)
	Mean (SD)	Median (Range)	Mean (SD)	Median (Range)	Mean (SD)	Median (Range)		
1 month	34 (14.6)^{B}	35.1 (13.8–57.4)	73.7 (25.5)^{A}	62.6 (44.9–100)	72.1 (31.1)^{A}	78.5 (27.2–100)	0.003*	0.382
3 months	73.8 (21)	69.5 (45.4–100)	90.6 (11.8)	100 (70.1–100)	93.1 (10.8)	100 (71.8–100)	0.077	0.092
6 months	96.2 (6.9)	100 (79–100)	98.6 (4.4)	100 (87–100)	100 (0)	100 (100–100)	0.096	0.073

*Significant at P ≤ 0.05, different superscripts in the same row indicate statistically significant difference between groups, PRF: Platelet-rich fibrin.

significantly higher median percentage reduction in lesions size than HA group. After 3 as well as 6 months; there was no statistically significant difference between median percentage decreases in lesions size in the three groups.

The present study agreed with a previous study by Basta *et al*. stated that using of PRF in conjunction with the synthetic bone graft enhances the healing of periapical intrabony defects after apicectomy [50]. Elbattawy *et al*. concluded that nHA produced a significant reduction in clinical and radiographic outcomes after 6 months in agreement with the present study [51].

Khetarpal *et al*. concluded that MTA in presence with PRF accelerates periapical healing in difficult cases as PRF is a strong fibrin membrane enriched with platelet and growth factors that accelerate periapical healing [52].

Conclusion

It was concluded that nHA combination with PRF produced faster periapical healing (bone regeneration) in the first 3 months than nHA alone. However, HA produces periapical healing (bone regeneration) after 6 months.

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