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TITLE: ENHANCING PERIAPICAL HEALING IN ENDODONTIC RETREATMENT CASES: A CLINICAL COMPARATIVE STUDY COMPARING NANO-HYDROXYAPATITE ALONE AND COMBINED WITH PRF

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ABSTRACT

Aim: This study aims to assess bone healing after periapical surgery using nHA and nHA+PRF.

Methods: Sixteen patients with periapical lesions in upper or lower anterior teeth with failed endodontic treatments were included. Teeth were divided into two groups of eight. All teeth were retreated in two visits. During the initial visit, the existing root canal filling was removed using ProTaper retreatment files (Dentsply Sirona®). Subsequently, the root canal was irrigated with a 2.5% sodium hypochlorite solution. Following irrigation, the canal was thoroughly dried and filled with a Bi-mix antibiotic paste composed of metronidazole and ciprofloxacin. During the second visit, canals were obturated with gutta-percha and sealer, followed by apicoectomy and root-end filling with MTA. In Group 1; nanohydroxyapatite (nHA) powder was packed into the bony periapical cavity. In Group 2, a mixture of nanohydroxyapatite (nHA) and platelet-rich fibrin (PRF) was packed into the bony periapical cavity. Follow-up visits were scheduled at 1, 3, and 6 months postoperatively for clinical and radiological assessments.

Results: At one month follow up, the nHA and nHA+PRF groups showed significantly greater reduction in lesion size. No significant differences were observed between the two groups either at three or six months.

Conclusion: nHA+PRF accelerated early bone regeneration compared to nHA alone

KEYWORDS: Periapical pathosis, bone healing, nano-hydroxyapatite,PRF ,retreatment

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INTRODUCTION

Microbial pulpal infections can results in the development of periapical lesions [1], like granulomas, cysts, or abscesses. [2,3] Successful root canal treatment depends on complete periapical healing, which is often achieved through non-surgical endodontic procedures. [4] Nevertheless, there are instances where symptoms persist, and infection remains despite non-surgical treatment. In such cases, periradicular surgery may be necessary to remove the pathological tissues, eliminating the irritation sources. This surgical intervention aims to address any residual infection and facilitate the regeneration of healthy periapical tissue[5]. Its widely recommended for bone regeneration and the promotion of soft tissue healing following oral surgery local utilization of growth factors, and plasma derivatives . One such approach involves the use of Platelet-Rich Fibrin (PRF), which has shown success when combined with osseus grafts in the treatment of periapical defects to regenerate the bone [6]. PRF stimulates a cascade of healing events, including cell proliferation, collagen synthesis, and angiogenesis, which contribute to enhanced tissue regeneration. When used in conjunction with bone grafts, PRF has been shown to result in excellent osseous defect fill, as evidenced by radiographic assessment, making it a valuable tool in accelerating bone healing and improving outcomes in periapical and other oral surgical procedures[6]. Various bone graft materials, including bioactive calcium phosphate ceramics, have been utilized to improve osseus healing. These materials, including hydroxyapatite (HA) and tricalcium phosphate (TCP), represent the largest family of alloplastic grafts. Both HA and TCP have been employed extensively to promote bone regeneration and enhance bone fill following periapical surgery. Their bioactive properties encourage osteo-conduction, allowing new bone to grow along the scaffold, and in some cases, they can even stimulate osteo-induction, further supporting the healing process. As a result, these calcium phosphate ceramics are valuable adjuncts in the treatment of periapical pathologies[7-10]. However, several studies had recommended using bone substitutes to pack the defect after surgery, with the goal of accelerating the healing process. They helped to restore the lost bone volume and provide a scaffold for new bone formation, facilitating faster regeneration and reducing the risk of complications. By providing structural support and promoting tissue repair and enhancing the outcomes of periradicular surgery[11-12]. Recently, nanotechnology has been introduced into dentistry to create and utilize materials, devices, and systems by controlling matter at the nanometer scale, which involves manipulating atoms, molecules, and supramolecular structures. This advanced approach allows for the development of dental materials with enhanced properties, such as improved strength, biocompatibility, and functional performance. By leveraging the unique characteristics of nanomaterials, such as their increased surface area and reactivity, it has the potential to significantly enhance overall clinical outcomes [13]. Nanohydroxyapatite (nHA) possesses distinctive characteristics due to its small size and large surface area, which enhances its interactions with biological tissues. In the present study, nHA was utilized alone and mixed with Platelet-Rich Fibrin (PRF), to promote osseus healing in the periapical area. Aiming to evaluate the potential advantages of using nHA and its combination with PRF in enhancing bone regeneration following periapical surgery.

PATIENTS AND METHODOLOGY

Sample Size Determination:

A power analysis was conducted to determine a sample size of 16 per group to detect a medium effect size with 80% power and a 5% significance level. [14,15].

Inclusion Criteria for Patient Selection:

Patient selection was based on the following standards:

1. **Amenability to engage in the study:** Patients should be willing to cooperate with the study and commit to attending regular follow-up visits throughout the study period.
2. **Systemic Health:** Patients should be free from chronic conditions that could affect anesthesia or negatively affect healing.
3. **Clinical Condition:** Patients must present with an anterior necrotic tooth exhibiting a periapical lesion measuring at least 5 millimeters in diameter, with a history of failed endodontic treatment.
4. **Informed Consent:** Patients provided informed consent before participating.
5. **Confidentiality:** All reasonable measures were taken to protect the privacy and security of the patients' personal and health information. All data was kept confidential, and the study was approved by the ethics committee.

These criteria ensured appropriate selection of participants while maintaining ethical standards and patient confidentiality throughout the study.

Criteria for exclusion:

1. **Chemotherapy or Radiotherapy**: Patients who received recent head and neck radiation or chemotherapy within the past year, as these treatments can impair healing and complicate the outcomes of the study.
2. **Medications Affecting Healing**: Patients taking medications that may negatively impact the healing process, such as systemic steroids or anticoagulant therapy, were excluded due to their potential to interfere with bone regeneration and tissue repair.
3. **Poor Oral Hygiene and Periodontal Issues**: Patients with poor oral hygiene or significant periodontal problems, as these conditions could affect the healing process and complicate the study's objectives.

Sixteen patients with periapical lesions were included in this study. Periapical radiographs and CBCT scans were used to assess the size of periapical lesions (≥5 mm) in failed endodontically treated single-rooted teeth. 16 teeth were randomly divided into two groups.

Clinical steps:

First visit: Preoperative radiographs were taken. Teeth were anesthetized (4% articaine local anesthesia), accessed, and isolated with a rubber dam. Teeth lengths were measured radiographically and electronically using apex locator (Mini Root ZX mini,J Morita, USA). Canals were cleaned and shaped with ProTaper files. Canals were irrigated with 2.5% NaOCl and dried. Bi-antibiotic paste(metronidazole and ciprofloxacin) was placed. Access cavities were sealed with composite resin. This initial procedure aimed to disinfect and temporarily seal the canals, allowing for subsequent interventions, and healing of the periapical tissues prior to the final filling and restoration phase.

Second Visit: (10 days later)

Patients were re-examined after 10 days. If asymptomatic, they rinsed with 0.2% chlorhexidine and proceeded to obturation and surgery. If symptomatic, canals were re-treated with NaOCl and bi-antibiotic paste for 4-6 weeks.

Periapical Surgery procedures:

Periapical surgery was performed under local anesthesia. A modified rectangular flap was elevated, apical curettage and resection of the root-end were performed. Root-end cavity was filled by MTA.

Group Assignments:

Group 1: Nanohydroxyapatite (nHA) powder was carefully condensed inside the bony cavity and to ensure entire packing of the void space and optimal contact with the surrounding bone tissue.

Group 2: **Preparation of Platelet-Rich Fibrin (PRF) Gel:** A volume of 10-30 mL of the patient's blood was collected into sterile, dry Monovettes without the addition of anticoagulants prior to the surgical procedure. The collected blood was immediately subjected to centrifugation at 2500 revolutions per minute for 10 minutes. This centrifugation process resulted in the separation of the blood components into 3 layers: RBC's, PRP, and a fibrin clot. The fibrin clot was carefully extracted using sterile forceps and transferred into a sterile tube [16] (Fig. 1). The nHA powder was added to the previously prepared platelet-rich fibrin (PRF) gel to mix them, then the mixture was condesed into the bony defect. (Fig. 2) followed by wound suturing. Instructions were given to the patients to apply cold compresses to the surgical site for 15 minutes every hour for the first three hours postoperatively.

Patients received antibiotics (Augmentin+Flagyl) and pain relievers (Bi-Profenid) for 5 days. Mouth rinses with warm saline and chlorhexidine were prescribed for 10 days. All sutures were removed after a period of 10 days. Patients were followed up at one, three and six months for clinical exams and radiographs [17].

Digital radiographs were taken using a size two sensor and film holders to allow for film positioning in a parallel position. After a period of six months, cone-beam computed tomography (CBCT) scans were obtained to assess bone density and healing. Linear dimensional measurements in millimiters were made on pre- and post-operative radiographs to evaluate bone defect reduction. [18-20].

Results data were non-normally distributed (Kolmogorov-Smirnov and Shapiro-Wilk tests). Data is presented as mean, SD, median, and range. Kruskal-Wallis and Friedman tests were used for between- and within-group comparisons, respectively. Dunn's test was used for post-hoc analysis. Statistical significance was set at $p<0.05$. Data were analyzed using IBM SPSS Statistics.

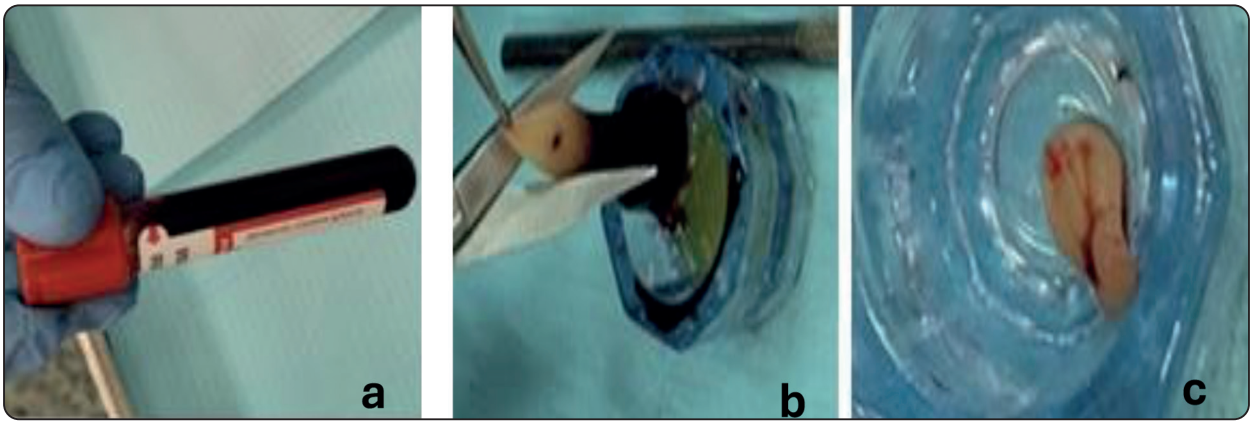

Fig. (1) Illustrating (a)10mL of blood was collected in a sterile tube, centrifuged, (b,c) the fibrin clot was extracted.

Fig. (2) illustrating PRF was mixed with nHA and packed into the bony cavity.

RESULTS

No significant differences in lesion size were found between groups preoperatively, 1, 3, and 6 months. (Tables 1-3)

- **Group 1 group showed** Significant reduction in lesion size was observed over time ($p < 0.001$). Pairwise comparisons showed significant decreases at 1, 1-3, and 3-6 months, suggesting a continuous and sustained healing process. (Fig. 3)
- **Group 2:** Significant reduction in lesion size over time ($p<0.001$). Significant decreases at 1 and 1-3 months, suggesting an initial rapid healing phase. However, no statistically significant change in lesion size was observed from 3 to 6 months, indicating that the healing process had stabilized (Fig. 4).

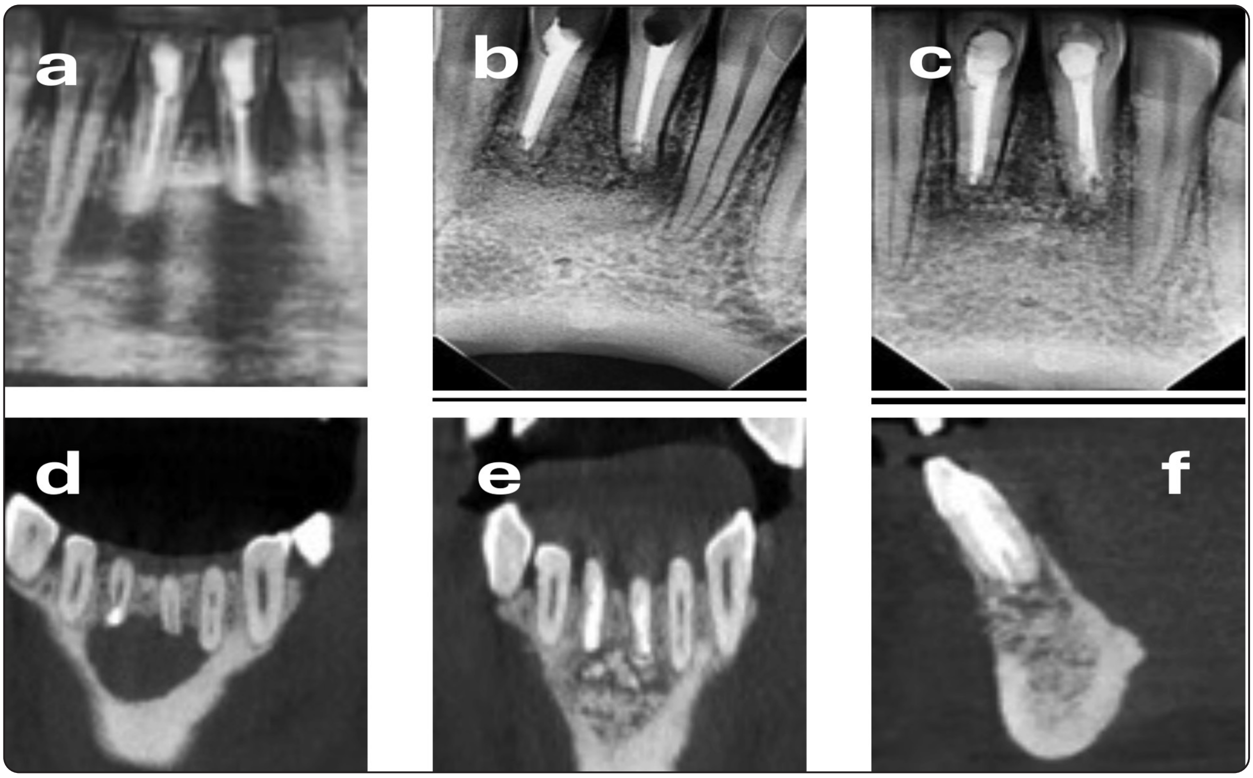

Fig. (3) illustrates nanohydroxyapatite group: a) preoperative radiograph for lower central incisors lesion: 11.5mm. b and c) post-surgical follow up at 3 and 6 months respectively. d) CBCT preoperative coronal view for lower central incisors e and f) CBCT 6 months coronal and sagittal view, respectively revealing complete bone healing.

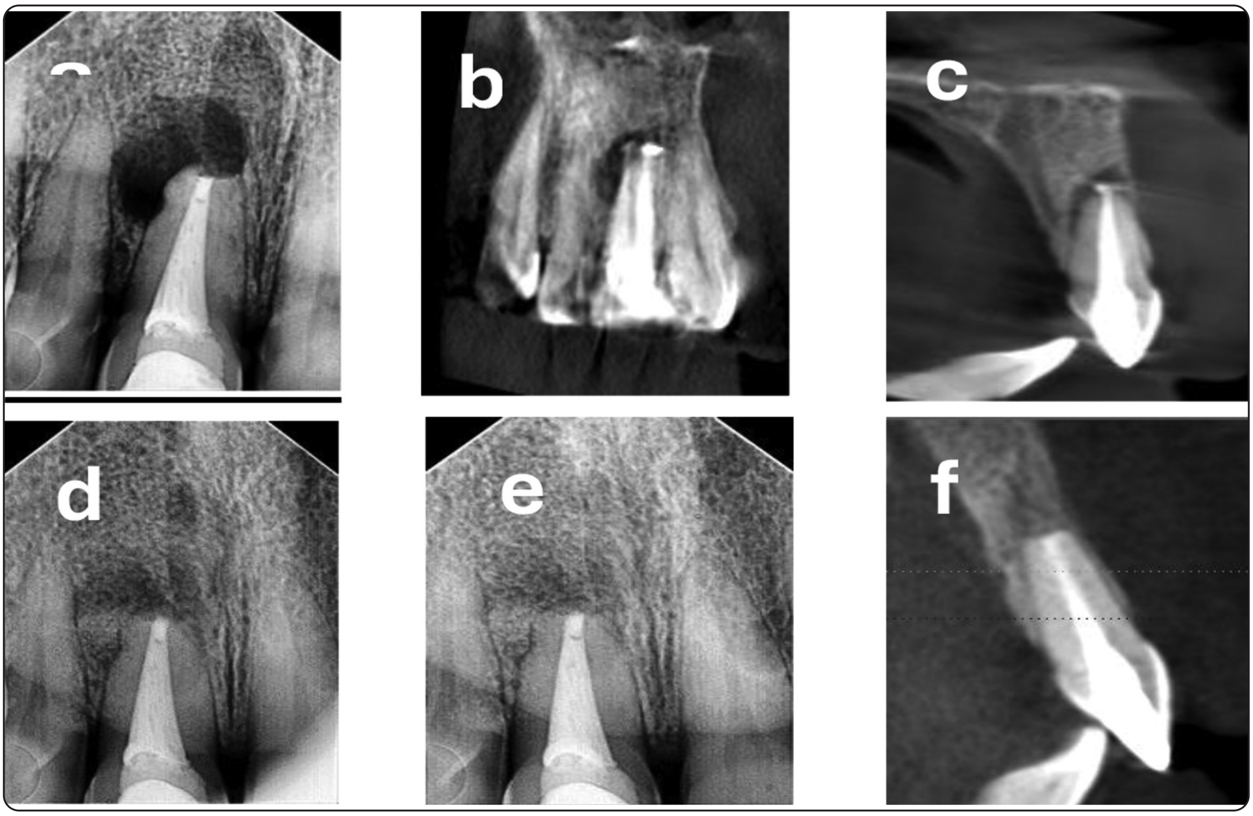

Fig. (4) illustrates nanohydroxyapatite and PRF group: a) post obturation for upper central lesion: 9.0 mm and before surgical procedure. b and c) presurgical CBCT Coronal and Sagittal view respectively. d and e) periapical radiograph after 3and 6 months follow up revealing bone healing. f) 6 months follow-up CBCT Sagittal view, revealing complete bone healing.

Percentage reduction in lesion size: calculating the percentage reduction in lesion size, it was determined as follows: [(Pre-operative size - Post-operative size) / Pre-operative size] x 100

The nano-HA group showed significantly greater reduction in lesions in 1 month compared to the PRF and nano-HA group (P = 0.003, Effect size = 0.382). During the three and six months, no significant differences were found in the median percentage decrease in lesion size across the two groups (P = 0.077, Effect size = 0.092; P = 0.096, Effect size = 0.073, respectively). (table 3)

TABLE (1). Kruskal-Wallis test results and descriptive statistics for lesion size comparison.

Time	Nano-Hydroxyapatite (Group 1)		PRF and Nano Hydroxyapatite (Group 2)		*P*-value	*Effect size (Eta Squared)*
	Mean (SD)	Median (Range)	Mean (SD)	Median (Range)		
Preoperative	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	3.5 (3.7)	4.1 (0-10.3)	2.4 (2.9)	1.8 (0-7.5)	0.122	0.053
3 months	1.3 (1.9)	0 (0-5.6)	0.7 (1.1)	0 (0-2.9)	0.205	0.007
6 months	0.2 (0.5)	0 (0-1.5)	0 (0)	0 (0-0)	0.096	0.073

TABLE (2). Illustrating descriptive statistics and results of Friedman's test for comparison of lesion size (mm) at different follow-up periods within each group.

Period	Group 1		Group 2	
	Mean (SD)	Median (Range)	Mean (SD)	Median (Range)
Pre- operative	10.2 (4.8) A	8.3 (5-18.7)	8.3 (2.4) A	8.1 (5.6-12.1)
1 month	3.5 (3.7) B	4.1 (0-10.3)	2.4 (2.9) B	1.8 (0-7.5)
3 months	1.3 (1.9) C	0 (0-5.6)	0.7 (1.1) C	0 (0-2.9)
6 months	0.2 (0.5) D	0 (0-1.5)	0 (0) C	0 (0-0)
P-value	<0.001*		<0.001*	
Effect size (w)	0.878		0.875	

TABLE (3) Illustrates the percentage reduction in lesion size.

Time	Nano-Hydroxyapatite		PRF and Nano- Hydroxyapatite		*P*-value	*Effect size (Eta Squared)*
	Mean (SD)	Median (Range)	Mean (SD)	Median (Range)		
1 month	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	90.6 (11.8)	100 (70.1-100)	93.1 (10.8)	100 (71.8-100)	0.077	0.092
6 months	98.6 (4.4)	100 (87-100)	100 (0)	100 (100- 100)	0.096	0.073

Note: the significance of the superscripts, such as: Different superscripts in the same row indicate statistically significant differences between groups at the corresponding time point.

DISCUSSION

Periapical surgery aims to regenerate lost bone tissue and restore oral health. However, inadequate bone healing, often caused by the ingrowth of non-mineralized tissue, can hinder the healing process. To facilitate bone regeneration, biocompatible materials, such as bone grafts or bone substitutes, can be used to fill the bony defect and provide a suitable environment for bone cells to proliferate and differentiate[21]. Ideal wound healing aims for maximum regeneration and minimal scarring. Tissue regeneration depends on the availability of cells and growth factors. Bioactive materials like hydroxyapatite (HA) can support bone regeneration[22]. This study compared nHA and nHA+PRF for periapical bone regeneration. Hydroxyapatite is a widely used bone graft material with similar structure to natural bone. Smaller particle size increases surface area, facilitating the absorption of essential proteins from blood plasma into the interstices, promoting cell growth. Ideal nano-bone graft materials should exhibit osteo-inductive properties, be fully synthetic, possess high porosity, possess a nanostructured architecture, have the ability to absorb protein particles within its nanoporous structure, and biodegradable by osteoclastic cells [23-27].

Nanohydroxyapatite (nHA), a synthetic bio-ceramic with a nanoscale structure, the small size and large surface area enhance bioreactivity[28]. Synthetic nHA bone grafts are widely used[29]. nHA's similarity to natural bone promotes osteoblast proliferation and metabolism, leading to better osseointegration and osteoconductivity. nHA can release calcium and phosphate ions, stimulating bone formation and inducing bone morphogenetic protein secretion, which help in the recruitment and differentiation of osteoprogenitor cells into mature osteoblasts, leading to the formation of new bone tissue [30-32].

Choukroun's PRF is an autologous platelet concentrate produced without anticoagulants, the lack of anticoagulants facilitates rapid platelets activation, triggering the coagulation leading to the formation of a fibrin clot. Platelets are activated naturally, releasing growth factors. PRF is a fibrin matrix that intrinsically embeds a large quantity of containing platelets and cytokines, which are released over time as the fibrin degrades, providing a sustained release of growth factors to the wound site[33-37].

In the present study, (PRF) gel was utilized as a bioactive scaffold to promote tissue regeneration. It is a natural, autologous biomaterial which is rich in growth factors and cytokines.[6] These growth factors promote wound healing and tissue regeneration. The IGF-1, in particular, has been shown to stimulate bone formation by promoting the proliferation and differentiation of osteoblasts which stimulate bone formation and cell proliferation[38-39].

The combination of nanohydroxyapatite and platelet-rich fibrin was employed in this study to synergistically enhance bone regeneration and minimize scar tissue formation. Many studies had observed that PRF combined with nanocrystalline HA and collagen can significantly enhance bone regeneration compared to conventional techniques and PRF alone[40-41].

Several factors contributed to the success of the surgical treatment in the current work, employing the crown-down technique. It facilitated direct access to the apical third, removal of large volume of necrotic materials and bacteria prior apical preparation, allowing for more penetration depth of the irrigants into the lateral canals, and facilitates control over the entire length of the canal. NaOCl was used as the disinfectant solution due to the antimicrobial properties along with tissue dissolution capabilities Furthermore, the attainment of an apical seal, achieved through the utilization of root-end filling materials, is essential to prevent continous ingress of bacteria and oral fluids into the periapical tissues, thereby promoting healing and preventing recurrent infection. Mineral Trioxide Aggregate (MTA) is widely considered the preferred retrograde filling material due to its superior sealing properties, biocompatibility, and high healing rates .[43-46]

A bimix antibiotic paste was utilized as an intracanal medicament between visits, which has been investigated as a potential therapeutic strategy for intracanal medicaments[47-49]. It was observed that nano-HA group showed significant reductions in lesion size over time. While, the PRF and nano-HA also showed significant reductions at 1 and 1-3 months, but not at 3-6 months.

Regarding the nano-HA+PRF and nano-HA groups at 1, 3, and 6 months. No significant difference was found between them. They showed significant reduction in lesion size. The present findings align with Basta et al., suggesting that combining PRF with synthetic bone grafts enhances periapical healing[50]. Elbattawy et al. found that nHA significantly reduced clinical and radiographic parameters after 6 months, aligning with our findings[51].

Khetarpal et al. demonstrated that the combination of MTA with platelets-rich fibrin can significantly promote the healing in complex cases. Since the PRF, a natural biological material from the patient's own blood, is enriched with a variety of growth factors, which played a crucial role in accelerating the healing process in the present study. By combining the biocompatibility and sealing properties of MTA with the regenerative potential of PRF, this synergistic approach lead to improved clinical outcomes and reduced healing time. [52]

RECOMMENDATIONS AND CONCLUSIONS

The results of this study exhibited that the combination of nanohydroxyapatite and platelet-rich fibrin significantly accelerated periapical healing within the first 3 months postoperatively compared to nHA alone. The synergistic effect of nHA and PRF, which involves the release of growth factors from PRF and the osteoconductive properties of nHA, appears to enhance tissue regeneration and accelerate bone formation.

Limitations of the study:

- The cost-effectiveness of the combination therapy compared to other treatment options should be evaluated. While the study demonstrates improved healing, it's important to consider the economic implications.
- The findings may not be directly applicable to all patients with periapical lesions. Factors such as patient age, systemic health, and the severity of the lesion can influence healing outcomes.

REFERENCES

1. Möller AJ, Fabricius L, Dahlén G, Ohman AE, Heyden G. Influence on periapical tissues of indigenous oral bacteria and necrotic pulp tissue in monkeys. Scand J Dent Res. 1981;89(6):475-84. https://doi.org/1111/j.1600-0722.1981.tb01711.x PMid:6951246
2. Bhaskar SN. Oral surgery oral pathology conference no. 17, walter reed army medical center. Periapical lesions--types, incidence, and clinical features. Oral Surg Oral Med Oral Pathol. 1966;21(5):657-71. https://doi.org/1016/0030-4220(66)90044-2 PMid:5218749
3. Lalonde ER, Luebke RG. The frequency and distribution of periapical cysts and granulomas. An evaluation of 800 specimens. Oral Surg Oral Med Oral Pathol. 1968;25(6):861-8. https://doi. org/1016/0030-4220(68)90163-1 PMid:5239741
4. Shah N. Nonsurgical management of periapical lesions: A prospective study. Oral Surg Oral Med Oral Pathol. 1988;66(3):365-71. https://doi. org/1016/0030-4220(88)90247-2 PMid:3174072
5. Torabinejad M, Corr R, Handysides R, Shabahang S. Outcomes of nonsurgical retreatment and endodontic surgery: A systematic review. J Endod. 2009;35(7):9307. https://doi.org/1016/j. joen.2009.04.023 PMid:19567310
6. Jayalakshmi KB, Agarwal S, Singh MP, Vishwanath BT, Krishna A, Agrawal R. Platelet-rich fibrin with β-tricalcium phosphate-A noval approach for bone augmentation in chronic periapical lesion: A case report. Case Rep Dent. 2012;2012:902858. https://doi.org/1155/2012/902858 PMid:23119189
7. Garrett K, Kerr M, Hartwell G, O'Sullivan S, Mayer P. The effect of a bioresorbable matrix barrier in endodontic surgery on the rate of periapical healing: An in vivo study. J Endod. 2002;28(7):503-6. https://doi.org/1097/00004770-200207000-00003 PMid:12126375
8. Harbert H. Generic tricalcium phosphate plugs: An adjunct in endodontics. J Endod. 1991;17(3):131-4. https://doi.org/1016/ S0099-2399(06)81746-2 PMid:1940729
9. Himel VT, Brady J Jr., Weir J Jr. Evaluation of repair of mechanical perforations of the pulp chamber floor using biodegradable tricalcium phosphate or calcium hydroxide. J Endod. 1985;11(4):161-5. https://doi.org/1016/ S0099-2399(85)80140-0 PMid:3858408
10. Jaber L, Mascrès C, Donohue WB. Reaction of the dental pulp to hydroxyapatite. Oral Surg Oral Med Oral Pathol. 1992;73(1):92 8. https://doi.org/1016/0030-4220(92)90162-j PMid:1318535
11. Lovelace TB, Mellonig JT, Meffert RM, Jones AA, Nummikoski PV, Cochran DL. Clinical evaluation of bioactive glass in the treatment of periodontal osseous defects in humans. J Periodontol. 1998;69(9):1027-35. https://doi.org/1902/ jop.1998.69.9.1027 PMid:9776031
12. Schwartz Z, Mellonig JT, Carnes DL Jr., de la Fontaine J, Cochran DL, Dean DD, et al. Ability of commercial demineralized freeze-dried bone allograft to induce new bone formation. J Periodontol. 1996;67(9):918-26. https://doi.org/1902/ jop.1996.67.9.918 PMid:8884650
13. Gholami GA, Najafi B, Mashhadiabbas F, Goetz W, Najafi S. Clinical, histologic and histomorphometric evaluation of socket preservation using a synthetic nanocrystalline hydroxyapatite in comparison with a bovine xenograft: A randomized clinical trial. Clin Oral Implants Res. 2012;23(10):1198-204. https://doi. org/1111/j.1600-0501.2011.02288.x PMid:22092485

14. Johns DA, Varughese JM, Thomas K, Abraham A, James EP, Maroli RK. Clinical and radiographical evaluation of the healing of large periapical lesions using triple antibiotic paste, photo activated disinfection and calcium hydroxide when used as root canal disinfectant. J Clin Exp Dent. 2014;6(3):e230.

15. Faul F, Erdfelder E, Lang AG, Buchner A. G* power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. Behav Res Methods. 2007; 39(2):175-91. https://doi.org/10.3758/bf03193146 PMid:17695343

16. Choukroun J, Adda F, Schoeffel C, Vervelle A. an opportunity in paro-implantology: PRF. Implantodontie. 2001;42:55-62.

17. Forsberg J, Halse A. Periapical radiolucencies as evaluated by bisecting-angle and paralleling radiographic techniques. Int Endod J. 1997;30(2):115-23. https://doi.org/1046/j.1365-2591.1997.00059.x PMid:10332245

18. Singh VP, Nayak DG, Uppoor AS, Shah D. Clinical and radiographic evaluation of Nano-crystalline hydroxyapatite bone graft (Sybograf) in combination with bioresorbable collagen membrane (Periocol) in periodontal intrabony defects. Dent Res J (Isfahan). 2012;9(1):60-7. https://doi.org/4103/1735-3327.92945 PMid:22363365

19. Kattimani VS, Chakravarthi PS, Kanumuru NR, Subbarao VV, Sidharthan A, Kumar TS, et al. Eggshell derived hydroxyapatite as bone graft substitute in the healing of maxillary cystic bone defects: A preliminary report. J Int Oral Health. 2014;6(3):15-9. PMid:25083027

20. Monga P, Grover R, Mahajan P, Keshav V, Singh N, Singh G. A comparative clinical study to evaluate the healing of large periapical lesions using platelet-rich fibrin and hydroxyapatite. Endodontology. 2016;28:27-31. https://doi.org/4103/0970-7212.184336

21. Sreedevi P, Varghese N, Varugheese JM. Prognosis of periapical surgery using bonegrafts: A clinical study. J Conserv Dent. 2011;14(1):68-72. https://doi.org/4103/0972-0707.80743 PMid:21691510

22. Goyal L. Clinical effectiveness of combining platelet rich f ibrin with alloplastic bone substitute for the management of combined endodontic periodontal lesion. Restor Dent Endod. 2014;39(1):51-5. https://doi.org/5395/rde.2014.39.1.51 PMid:24516830

23. Lee JS, Park WY, Cha JK, Jung UW, Kim CS, Lee YK, et al. Periodontal tissue reaction to customized nano-hydroxyapatite block scaffold in one-wall intrabony defect: A histologic study in dogs. J Periodontal Implant Sci. 2012;42(2):50-8. https://doi. org/5051/jpis.2012.42.2.50 PMid:22586523

24. Fathi MH, Mortazavi V, Esfahani SI. Bioactivity evaluation of synthetic nanocrystalline hydroxyapatite. Dent Res J. 2008;5:81-7.

25. Mantri SS, Mantri SP. The nano era in dentistry. J Nat Sci Biol Med. 2013;4(1):39-44. https://doi.org/4103/0976-9668.107258 PMid:23633833

26. Liu H, Webster TJ. Nanomedicine for implants: A review of studies and necessary experimental tools. Biomaterials. 2007;28(2):354 69. https://doi.org/1016/j.biomaterials.2006.08.049 PMid:21898921

27. Wasem M, Köser J, Hess S, Gnecco E, Meyer E. Exploring the retention properties of CaF2 nanoparticles as possible additives for dental care application with tapping-mode atomic force microscope in liquid. Beilstein J. Nanotechnol. 2014;5:36-43. https://doi.org/10.3762/bjnano.5.4

28. Jahangirnezhad M, Kazeminezhad I, Saki G, Amirpoor S, Larki M. The effects of Nanohydroxyapatite on bone regeneration in rat calvarial defects. Am J Res Commun. 2013;1(4):302-16.

29. Zhou H, Lee J. Nanoscale hydroxyapatite particles for bone tissue engineering. Acta Biomater. 2011;7(7):2769-81. https://doi.org/10.1016/j.actbio.2011.03.019 PMid:21440094

30. Barkarmo S, Wennerberg A, Hoffman M, Kjellin P, Breding K, Handa P, et al. Nano-hydroxyapatite-coated PEEK implants: A pilot study in rabbit bone. J Biomed Mater Res A. 2013;101(2):465-71. https://doi.org/1002/jbm.a.34358 PMid:22865597

31. Hu J, Zhou Y, Huang L, Liu J, Lu H. Effect of nano hydroxyapatite coating on the osteoinductivity of porous biphasic calcium phosphate ceramics. BMC Musculoskelet Disord. 2014;1(15):114. https://doi.org/1186/1471-2474-15-114 PMid:24690170

32. Pilloni A, Pompa G, Saccucci M, Di Carlo G, Rimondini L, Brama M, et al. Analysis of human alveolar osteoblast behavior on a nano-hydroxyapatite substrate: An in vitro study. BMC Oral Health. 2014;14:22. https://doi.org/1186/1472-6831-14-22

33. Shivashankar VY, Johns DA, Vidyanath S, Sam G. Combination of platelet rich fibrin, hydroxyapatite and PRF membrane in the management of large inflammatory

periapical lesion. J Conserv Dent. 2013;16(3):261-4. https://doi.org/4103/0972-0707.111329 PMid:23833463

34. Von Arx T, Alsaeed M. The use of regenerative techniques in apical surgery: A literature review. Saudi Dent J. 2011;23(3):113-27. https://doi.org/1016/j.sdentj.2011.02.004 PMid:24151420

35. Tsesis I, Rosen E, Tamse A, Taschieri S, Del Fabbro M. Effect of guided tissue regeneration on the outcome of surgical endodontic treatment: A systematic review and meta-analysis. J Endod. 2011;37(8):1039-45. https://doi.org/1016/j.joen.2011.05.016 PMid:21763891

36. Bashutski JD, Wang HL. Periodontal and endodontic regeneration. J Endod. 2009;35(3):321-8. https://doi.org/1016/j.joen.2008.11.023 PMid:19249588

37. He L, Lin Y, Hu X, Zhang Y, Wu H. A comparative study of platelet-rich fibrin (PRF) and platelet-rich plasma (PRP) on the effect of proliferation and differentiation of rat osteoblasts in vitro. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2009;108(5):707-13. https://doi.org/1016/j.tripleo.2009.06.044 PMid:19836723

38. Shimao Y, Mingguo W, Jing L, Jinpan L, Xialian L, Wei X. The comparison of platelet-rich fîbrin and platelet-rich plasma in releasing of growth factors and their effects on the proliferation and differentiation of adipose tissue derived stem cells in vitro. Hua Xi Kou Qiang Yi Xue Za Zhi 2012;30:6.

39. Singh S, Singh A, Singh S, Singh R. Application of PRF in surgical management of periapical lesions. Natl J Maxillofac Surg. 2013;4(1):94-9. https://doi.org/4103/0975-5950.117825 PMid:24163562

40. Thanikasalam M, Ahamed S, Narayana SS, Bhavani S, Rajaraman G. Evaluation of healing after periapical surgery using platelet-rich fibrin and nanocrystalline hydroxyapatite with collagen in combination with platelet-rich fibrin. Endodontology. 2018;30:25-31.

41. Abo Shady TE, Elgendy EA. Clinical and radiographic evaluation of nanocrystalline hydroxyapatite with or without platelet-rich f ibrin membrane in the treatment of periodontal intrabony defects. J Indian Soc Periodontol. 2015;19(1):61-5. PMid:25810595

42. Johnson BR. Considerations in the selection of a root end filling material. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 1999;87(4):398-404. https://doi.org/1016/s1079-2104(99)70237-4 PMid:10225620

43. Von Arx T, Peñarrocha M, Jensen S. Prognostic factors in apical surgery with root-end filling: A meta-analysis. J Endod. 2010;36(6):957-73. https://doi.org/1016/j.joen.2010.02.026 PMid:20478447

44. Eliyas S, Vere J, Ali Z, Harris I. Micro-surgical endodontics. Br Dent J. 2014;216(4):169-77. https://doi.org/1038/sj.bdj.2014.142 PMid:24557386

45. Carrotte P. Endodontics: Part 7. Preparing the root canal. Br Dent J. 2004;27:197(10):603-13. https://doi.org/1038/sj.bdj.4811823 PMid:15611742

46. Maity I, Meena N, Kumari RA. Single visit nonsurgical endodontic therapy for periapical cysts: A clinical study. Contemp Clin Dent. 2014;5(2):195-202. https://doi.org/4103/0976-237X.132321 PMid:24963246

47. Fouad AF. The microbial challenge to pulp regeneration. Adv Dent Res. 2011;23(3):285-9. https://doi.org/1177/0022034511405388 PMid:21677080

48. Nagata JY, Soares AJ, Souza-Filho FJ, Zaia AA, Ferraz CC, Almeida JF, et al. Microbial evaluation of traumatized teeth treated with triple antibiotic paste or calcium hydroxide with 2% chlorhexidine gel in pulp revascularization. J Endod. 2014;40(6):778-83. https://doi.org/1016/j.joen.2014.01.038 PMid:24862703

49. Sabrah AH, Yassen GH, Spolnik KJ, Hara AT, Platt JA, Gregory RL. Evaluation of residual antibacterial effect of human radicular dentin treated with triple and double antibiotic pastes. J Endod. 2015;41(7):1081-4. https://doi.org/1016/j.joen.2015.03.001 PMid:25887806

50. Basta DG, Abu-Seida AM, El-Batouty KM, Tawfik HM. Effect of combining platelet-rich fibrin with synthetic bone graft on the healing of intrabony defects after apicectomy in dogs with periapical pathosis. Saudi Endod J. 2021;11(3):300-7.

51. Elbattawy W, Ahmed D. Clinical and radiographic evaluation of open flap debridement with or without nanocrystalline hydroxyapatite bone graft in management of periodontal intrabony defects. Egypt Dent J. 2021;67:433-46.

52. Khetarpal A, Chaudhry S, Talwar S, Verma M. Endodontic management of open apex using MTA and platelet rich fibrin membrane barrier: A newer matrix concept. J Clin Exp Dent. 2013;5(5):e291-4. https://doi.org/10.4317/jced.51178 PMid:24455097