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CBCT-based comparative evaluation of A-PRF and i-PRF for periodontal regeneration in intrabony defects

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Abstract:
To optimize tissue healing, researchers have developed advanced platelet rich fibrin (A-PRF) and injectable platelet rich fibrin (i-PRF) by modifying traditional platelet rich fibrin (PRF) protocols to enhance soft and hard tissue regeneration. Therefore, it is of interest to evaluate how combining different PRF modalities with nanocrystalline hydroxyapatite bone graft effect the regeneration in 12 patients with bilateral intrabony defects. Clinical parameters were assessed at baseline, 3 and 6 months with CBCT evaluation performed at baseline and 6 months. Both treatment modalities resulted in significant clinical and radiographic improvements, with i-PRF showing superior soft tissue healing and enhanced graft handling characteristics. Thus, data shows that i-PRF provides practical surgical advantages while achieving regenerative outcomes comparable to A-PRF, thereby advancing its clinical application in periodontal intrabony defect management.

Keywords: Advanced platelet rich fibrin (A-PRF), injectable platelet rich fibrin (I-PRF), intrabony defect, periodontal regeneration, nanocrystalline hydroxyapatite (Nc-HA) bone graft

Background:
The periodontal therapy aims to halt disease process while restoring the structural and functional integrity of compromised tissues. Conventional surgical procedures have limited regenerative potential; therefore, successful periodontal regeneration depends on controlled progenitor cell migration, proliferation and repopulation within the defect to achieve functional re-establishment of the periodontal attachment apparatus **[1].** Although conventional periodontal surgery remains the standard therapeutic approach, its regenerative potential is limited. Advances in nano biomaterials, particularly nanocrystalline hydroxyapatite, have shown promising regenerative capabilities due to superior physicochemical and biological properties. Compared with microcrystalline forms, nanocrystalline hydroxyapatite enhances osteoblast adhesion, proliferation, differentiation and mineralization, thereby promoting accelerated and more predictable periodontal tissue regeneration with reduced healing duration **[2, 3]**. Recent research has progressively shifted towards growth factor mediated approaches to regulate and accelerate periodontal regeneration **[4]**. To maximize regenerative capacity Bajaj *et al.* (2017), Ghannati *et al* (2014) and Clark *et al.* (2018) modified traditional processing protocol to develop advanced platelet-rich fibrin (A-PRF), expanding therapeutic potential of platelet concentrates. A-PRF prepared using low-speed centrifugation (1500 rpm for 14 minutes), producing a highly elastic fibrin matrix enriched with neutrophils, granulocytes, monocytes, hematopoietic stem cells and T and B lymphocytes. These cellular components are uniformly distributed throughout the clot, particularly in the distal region, contributing significantly to bone regeneration. Morphologically, A-PRF shows loose fibrin architecture with enlarged interfibrous spaces, while immunohistochemical analyses confirm abundant $CD61^+$ platelets and extensive $CD15^+$ neutrophilic granulocyte distribution **[5-7]**. The Low-speed centrifugation protocol (700 rpm for 3 minutes) established by Miron *et al.* in 2015 yielded a liquid formula of platelet rich fibrin (i PRF), represents a significant advancement in platelet concentrate technology. Unlike solid PRF formulations, i PRF exist in liquid form, enabling easy combination with bone graft materials. Following coagulation, it forms a fibrin network that encapsulates graft particles, creating "sticky bone" with improved stability and handling properties. These fibrin–graft composite acts as a sustained reservoir of growth factors, promoting cellular proliferation and differentiation, thereby enhancing graft integration and facilitating more predictable periodontal regenerative outcomes **[8]**. Therefore, it is of interest to compare the clinical and radiographic outcomes of advanced platelet-rich fibrin (A-PRF) and injectable platelet-rich fibrin (i-PRF), each combined with nanocrystalline hydroxyapatite (Nc-HA), in the regenerative management of periodontal intrabony defects.

Materials and Methods:

Study design and sample size calculation:
This study strictly adhered to the Declaration of Helsinki principles and approval from Institutional Ethics Committee (MDC/IEC/2024/353 I). Before any procedures began, all patients were fully briefed on the study and provided written informed consent. G*Power 3.1 software was utilized for sample size estimation, setting the statistical power at 96% and the significance level (α) of 0.05; dictated a minimum of **8 defects per group**. To safeguard against potential patient dropouts, 12 patients were enrolled exhibiting one pair of vertical bone defect. The site was randomly assigned into two groups:

[1] Group I: A PRF + Nanocrystalline Hydroxyapatite bone graft.
[2] Group II: I PRF+ Nanocrystalline Hydroxyapatite bone graft.

Participants:
The study population includes systemically healthy male and female patients, aged 25-60 years, exhibiting two nonadjacent teeth showing pocket probing depths of 5 mm or more following initial phase I periodontal therapy, along with clear radiographic

signs of vertical bone loss. Conversely, we excluded individuals who had received periodontal treatment within past 6 months, those with poor oral hygiene compliance and pregnant or lactating women.

Advanced platelet-rich fibrin (A-PRF):
Prepared following Choukroun established technique 10 millilitres of intravenous blood, collected in sterile glass tubes. Samples were immediately centrifuged at 1500 rpm for 14 minutes, yielding a dense fibrin clot in the middle layer. We carefully extracted and separated the clot from red blood cell fraction, sectioned and mixed it with bone graft in a standardised 1:1 ratio **[7]**.

Injectable platelet-rich fibrin (I-PRF):
Prepared according to the protocol described by Miron *et al.* 5 millilitres of intravenous blood collected in a Vacutainer. Immediately centrifuged at 700 rpm for 3 minutes. The I-PRF layer was aspirated and combined with bone graft material within 10–15 minutes, allowing it to set into a cohesive, homogeneous graft mass **[8]**.

Surgical procedure:
All surgical interventions were performed under aseptic conditions using 2% lignocaine hydrochloride with 1:80,000 adrenalines. An intracrevicular incision was made with a Bard–Parker handle and blade and full-thickness mucoperiosteal flaps were elevated buccal and lingual/palatal to expose the intrabony defect. All granulation tissue was meticulously removed to obtain a biologically clean root surface. Defect morphology, including depth and residual osseous walls, was assessed intraoperatively using a calibrated periodontal probe with customized acrylic stents to ensure reproducibility of measurements. Group I: The A-PRF clot was combined with nanocrystalline hydroxyapatite and packed into the defect. Group II: the I-PRF was mixed with nanocrystalline hydroxyapatite and placed into the defect. Flaps were repositioned for tension-free primary closure and secured with interrupted 3-0 non-resorbable sutures. A periodontal dressing was applied. Postoperative care included systemic antibiotics, analgesics and 0.12% chlorhexidine rinses twice daily for 14 days. Periodontal dressing and sutures were removed after 10 days. Clinical parameters, including Plaque Index (PI), Gingival Index (GI), Probing Depth (PD) and Relative Attachment Level (RAL), were tracked at baseline, 3 months and 6 months post-operatively **[9]**. Cone-beam computed tomography (CBCT) scan was taken at baseline and 6 months postoperatively to evaluate changes in defect depth, bone fill and bone fill %. The base of the defect (BD) - distance from cemento-enamel junction (CEJ) to the most in-depth extension of the bony defect. Bone fill % = Bone defect at baseline-Bone defect at 6 months x 100 / bone defect at baseline **[10].**

Statistical analysis:
Managed via Statistical Package of Social Sciences (SPSS) software, v.22. Mean variable was compared between the treatment group across different intervals using independent Student t-test and ANOVA tests.

Results:
Every participant successfully completed the study with zero post-operative complications. Baseline clinical and radiographic data were comparable between both arms ($p > 0.05$), confirming effective randomization. Over six months, both groups showed significant clinical progress. Plaque Index (PI) scores decreased significantly for all participants ($p < 0.001$). Group I dropped from 1.00 ± 0.08 at baseline to 0.06 ± 0.08, while Group II decreased from 1.05 ± 0.13 to 0.45 ± 0.06, with Group II showing significantly greater intermediate and final improvements ($p < 0.001$). Gingival Index (GI) scores also fell: Group I went from 1.10 ± 0.14 to 0.75 ± 0.06, whereas Group II dropped from 1.13 ± 0.10 to 0.48 ± 0.10, heavily favoring Group II at both intervals ($p < 0.05$). Probing pocket depth (PPD) reductions were highly significant for both therapies ($p < 0.01$). Group I PPD decreased from 7.25 ± 0.96 mm to 4.00 ± 0.82 mm, while Group II saw a superior drop from 7.75 ± 0.96 mm to 2.00 ± 0.00 mm ($p < 0.001$). Relative attachment level (RAL) matched this trend ($p < 0.001$): Group I improved from10.25 ± 0.50 mm to 6.25 ± 0.50 mm, whereas Group II achieved greater gains, moving from 9.75 ± 0.96 mm to 4.50 ± 1.29 mm ($p < 0.001$) **(Table 1).** Radiographically, both cohorts achieved significant defect fill ($p < 0.001$). However, intergroup differences in bone fill percentage and defect depth reduction were not statistically significant ($p > 0.05$). Ultimately, while both modalities are effective, Group II (I-PRF) provides superior soft-tissue healing **(Table 2).**

Table 1: Comparison of the PI, GI, PPD and RAL at different timelines

Group	Timeline	Plaque Index		Mean Difference	F value	P value
		Mean	Standard Deviation			
Group I	Baseline	1.00	0.08	-	21.4	<0.001**
	3 months	0.83	0.10	0.175		
	6 months	0.60	0.08	0.4		
Group II	Baseline	1.05	0.13	-	37.3	<0.001**
	3 months	0.65	0.10	0.4		
	6 months	0.45	0.06	0.6		
		Gingival index				
Group I	Baseline	1.10	0.14	-	9.6	0.006*
	3 months	0.88	0.13	0.225		
	6 months	0.75	0.6	0.35		
Group II	Baseline	1.13	0.10	-	36.5	<0.001**
	3 months	0.75	0.13	0.375		
	6 months	0.48	0.10	0.65		
		PPD				

Group I	Baseline	7.25	0.96	-	8.2	0.009*
	3 months	5.75	1.50	1.5		
	6 months	4.00	0.82	3.25		
Group II	Baseline	7.75	0.96	-	94.1	<0.001**
	3 months	3.25	0.50	4.5		
	6 months	2.00	00	5.75		
			RAL			
Group I	Baseline	10.25	0.50	-	57.9	<0.001**
	3 months	8.50	0.58	1.75		
	6 months	6.25	0.50	5		
Group II	Baseline	9.75	0.96	-	15.4	<0.001**
	3 months	6.25	1.71	3.5		
	6 months	4.50	1.29	5.25		

*Repeated measures ANOVA, ** Highly Significant, * Significant*

Table 2: Comparison of Defect Depth, Bone fill & Bone fill % at 6-month time intervals

Group	Timeline	Defect Depth		Mean Difference	t value	P value
		Mean	Standard Deviation			
Group I	Baseline	5.75	0.83	-	7.08	<0.001**
	6 months	3.14	0.93	2.52		
Group II	Baseline	5.58	0.73	-	8.15	<0.001**
	6 months	2.8	0.93	2.58		
			Bone fill			
Group I	6 months	2.6	0.93	-	0.43	0.67#
Group II	6 months	2.78	1.09	2.52		
			Bone fill %			
Group I	6 months	50.08 %	17.16	-	0.37	0.97#
Group II	6 months	50.25 %	17.27	2.58		

*Student t test, ** highly significant, # non-significant.*

Discussion:

The purpose of regenerative periodontal therapy is to rebuild damaged periodontium. To enhance these clinical outcomes, autologous platelets concentrates have emerged as vital bioactive essential growth factors and accelerated tissue healing. A-PRF and i PRF utilize low speed centrifugation protocol to maximize cellular yield and optimize growth factor release **[7, 10]**. Nanocrystalline hydroxyapatite (Nc-HA), an osteoconductive biomaterial, frequently utilized to manage periodontal intrabony defects. Due to its structural characteristics mimicking natural bone mineral, NcHA serves as an effective three-dimensional scaffold that supports and guides proliferation of the new bone tissue **[11]**. These investigations evaluate the regenerative outcome of treating periodontal intrabony defect using combination of A-PRF + NcHA (Group I) against i-PRF + NcHA (Group II). Within each group, significant improvements were observed in plaque index (PI), gingival index (GI), probing depth (PD) and relative attachment level (RAL) from baseline to 3 and 6 months, alongside a notable reduction in radiographic defect depth. While intergroup analysis showed that Group II achieved significantly greater improvements in clinical parameters, no statistical difference in radiographic bone fill was detected between the two cohorts. Both groups achieved significant clinical success due to the biological activity of platelet concentrates. By acting as growth factor reservoirs (PDGF, TGF-β, VEGF, IGF), they accelerate periodontal healing **[6]**. The enhanced clinical results demonstrated by group II may be explained by its injectable liquid formulation, which enables uniform distribution within the defect and intimate mixing with particulate Nc-HA graft. Once it polymerizes, i-PRF creates a supportive fibrin mesh that locks in cells and growth factors, which helps stabilize the early wound and guides cells to where they need to go. This aligns with findings published by Miron *et al.* in 2017, which showed that i-PRF boasts a richer concentration of regenerative cells and growth factors than older PRF formulations **[8]**. Furthermore, Kannan *et al.* in 2025 noted that this injectable variant significantly boosts soft tissue repair and accelerates early clinical attachment gains **[12]**. In contrast, A-PRF develops a more durable fibrin matrix that slowly and steadily releases growth factors over an extended period **[6]**. Although this property supports regeneration, its solid architecture may limit homogeneous integration with particulate graft materials, which could explain why group I showed slight smaller improvement in probing depth and clinical attachment level. Nevertheless, both A-PRF and i-PRF clearly boosted overall clinical success when combined with Nc-HA. Despite superior clinical outcomes in Group II, radiographic bone fill did not differ significantly between groups, suggesting that hard tissue regeneration may be primarily governed by the osteoconductive properties of Nc-HA. Nanocrystalline hydroxyapatite is characterized by high surface area, nanocrystalline architecture and enhanced bioactivity, facilitating osteoblast adhesion and proliferation **[13]**. The research group led by Kasaj *et al.* (2008) and Schwarz *et al.* (2006) observed marked radiographic and clinical healing when treating intrabony defects using Nc-HA **[13, 14]**. Furthermore, randomized clinical trials have showed greater bone fill when platelet concentrates are combined with bone grafts compared to their use alone. Pradeep *et al.* (2012) reported mean bone fill of 56.46% ± 9.26% with PRF alone versus 63.39% ± 16.52% when combined with hydroxyapatite **[15]**. Recent research heavily favors mixing platelet concentrates with hydroxyapatite grafts for better regenerative results. Mallappa's *et al.* (2022) leveraged an advanced A-PRF block formulation to unlock superior pocket depth reductions, attachment gains and greater bone volume **[16]**. Mirroring these positive results, independent studies by Nair's *et al.* in 2022 and Chaudhary *et al.* and associates in 2023 confirmed that simply combining i-PRF with nHA or standard

hydroxyapatite significantly boosts healing outcomes **[17, 18]**. The LSCC-based preparation of A-PRF preserves higher cellular content and enables sustained release of bioactive mediators, whereas i-PRF, as a liquid concentrate, facilitates rapid polymerization into a cohesive graft matrix, improving handling and early angiogenesis **[6, 10 and 17]**. Studies by Rithesh *et al.* and Salama *et al.* (2025) have shown significantly enhanced radiographic bone fill and clinical attachment gain with i-PRF combined with graft materials **[19, 20]**. Evidence supporting A-PRF is also substantial, with Fujioka-Kobayashi *et al.* (2017), reporting favourable regenerative outcomes due to sustained growth factor release and immunomodulatory effects **[21]**. A recent goat metacarpal defect micro-CT study showed that i-PRF combined with β-tricalcium phosphate (β-TCP) produced significantly greater bone volume, mineral density and trabecular quality compared with PRF/β-TCP and β-TCP alone, indicating superior regenerative potential **[22]**. Similarly, when mixed with nano-crystalline hydroxyapatite (Nc-HA), both i-PRF and A-PRF may enhance osteogenesis and graft integration. These findings align with systematic reviews by Pullishery *et al.* (2024) which concluded that while platelet concentrates improve clinical parameters, radiographic bone gain between different PRF formulations often shows minimal differences **[23]**. Consistent with the present findings, Khan *et al.* (2026) showed significant clinical and radiographic improvements with both i-PRF and CGF **[24],** whereas Genescia *et al.* (2026) observed superior outcomes with A-PRF block therapy **[25].** These differences may reflect variations in fibrin architecture, cellular retention, growth-factor release, and biomaterial interaction, although direct comparisons remain constrained. Within the limitations of this study, including short follow-up duration, lack of histological validation and inter-individual variability in healing, both the groups combined with Nc-HA were effective in managing intrabony defects. However, i-PRF showed greater clinical outcomes, particularly in terms of reduced probing depth and attachment gain. Since radiographic bone fill was comparable, both modalities remain clinically viable and the choice may depend on handling characteristics, cost-effectiveness and operator preference. Moving forward the field would benefit most from large scale, multi-clinic randomized trial with extended follow up period to confirm if these initial healing trends hold up over time.

Conclusion:

Both A-PRF and i-PRF, when combined with nanocrystalline hydroxyapatite (Nc-HA), showed significant clinical and radiographical defect resolution. The i-PRF + Nc-HA group exhibited superior clinical parameters, indicating enhanced soft tissue healing and early wound stability. However, radiographic bone fill was comparable between groups, suggesting that both modalities are clinically effective, with i-PRF offering additional advantages in soft tissue outcomes.

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