

Molecular and Cellular Mechanisms of Platelet-Rich Fibrin in Dental Implant Osseointegration: Emerging Roles of MicroRNAs, Exosomes, and Mesenchymal Stem Cells

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Abstract

Background: Platelet-rich fibrin (PRF) is widely used as an autologous adjunct in implant dentistry, yet its biological action is usually attributed generically to growth-factor release. Three further regulatory layers (microRNAs, extracellular vesicles including exosomes, and mesenchymal stem cells) have recently been proposed as mediators of its effect on peri-implant bone healing.

Materials and Methods: Narrative review of PubMed/MEDLINE literature combining PRF formulations (L-PRF, A-PRF, i-PRF) with microRNA, exosome/extracellular vesicle, mesenchymal stem cell, macrophage polarization and osseointegration terms. Each finding was graded by model (in vitro, animal, human), and human implant studies were used to test mechanistic claims against clinical outcomes.

Results: PRF promotes proliferation, migration and osteogenic commitment of mesenchymal stem cells through ERK1/2, TGF- β /Smad2/3, PDGF/Akt and BMP2/Smad1/5/8 signalling, in a dose-dependent manner in which high i-PRF concentrations can impair osteogenic induction. PRF contains stable microRNAs (miR-16, miR-21, miR-146a, miR-155); L-PRF restored miR-21 in diabetic bone defects and i-PRF scaffolds drove osteogenesis through miR-21/miR-125a-mediated TGF- β /Smad signalling. PRF-derived extracellular vesicles carry TGF- β 1, VEGF, PDGF-A, miR-21 and miR-146a, and fibrin matrices can retain and release stem-cell vesicles for up to four weeks. Macrophage responses to PRF are time- and model-dependent. Almost all mechanistic data are preclinical or from non-oral tissues, whereas human implant studies show mainly transient early gains in implant stability and have not measured any molecular endpoint.

Conclusion: MicroRNAs, extracellular vesicles and mesenchymal stem cells provide a coherent biological rationale for PRF in osseointegration but not yet a validated clinical explanation. Protocol-stratified characterization of human PRF, dose-response studies and clinical trials with molecular endpoints are required.

Key Word: Platelet-rich fibrin; Osseointegration; MicroRNAs; Exosomes; Extracellular vesicles; Mesenchymal stem cells; Dental implants; Osteoimmunology.

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I. Introduction

Osseointegration is no longer described simply as bone growing against titanium. It is better understood as a sequence of cell-to-cell conversations that unfold across four overlapping phases (haemostasis, inflammation, proliferation and remodelling), each driven by cytokines, extracellular matrix proteins and small signalling molecules; when that sequence stalls in the early inflammatory phase, a toxic wound environment rich in neutrophils, radicals and proteases replaces the regenerative one [1]. The blood clot that forms at the bone-implant interface in the first minutes after insertion is therefore not a passive filler but the first scaffold, and the first signalling hub, of the entire healing cascade.

Platelet-rich fibrin (PRF) was designed to intervene precisely in that window. Obtained by centrifuging whole blood without anticoagulants, PRF polymerizes into a dense fibrin network that entraps platelets, leukocytes and their secretory products [2], and its liquid, injectable formulation (i-PRF) has since extended the concept to irrigation, surface coating and injection protocols [3]. In a recent critical review of PRF in implant dentistry, our group concluded that the clinical evidence is uneven across indications: comparatively stronger for soft-tissue healing and peri-implant biomodulation, more mixed for PRF as a stand-alone bone-graft substitute, and still emerging for newer non-surgical applications such as i-PRF in peri-implant disease [4]. Systematic appraisals of implant outcomes point in the same direction, with moderate evidence limited to the early phase of osseointegration [5], a secondary-stability meta-analysis of 12 trials, nine of them at high or unclear risk of bias,

yielding only low-certainty evidence [6], and a 2026 scoping review of 19 human studies that found mostly transient early gains in implant stability without durable superiority in marginal bone or survival [7].

A recurring weakness of this literature is mechanistic: PRF's effects are usually attributed, almost by default, to "growth factors" (PDGF, TGF- β , VEGF, IGF) released from the fibrin matrix. Over the past five years, three additional regulatory layers have come into focus. The first is post-transcriptional control by microRNAs (miRNAs), short non-coding RNAs that tune the translation of osteogenic, angiogenic and inflammatory genes. The second is intercellular transfer of cargo by extracellular vesicles (EVs), including the endosome-derived subset commonly called exosomes, which has led some authors to propose EV-based products as a third generation of platelet concentrates [8]. The third is the behaviour of the target cells themselves, particularly mesenchymal stem/stromal cells (MSCs), whose recruitment and lineage commitment ultimately determine whether bone forms on the implant surface.

The objective of this narrative review is to synthesize the current evidence on these three layers as they relate to PRF and dental implant osseointegration, to grade that evidence explicitly by model (in vitro, animal, human), and to identify where mechanistic understanding has outrun clinical validation. The review is written as a direct continuation of our earlier clinical synthesis [4], moving from the question "does PRF work?" to the more precise question "through which molecular and cellular pathways could it work, and what would be needed to prove it?".

II. Materials and Methods

This is a narrative (non-systematic) literature review. PubMed/MEDLINE was searched in September 2026 using combinations of "platelet-rich fibrin", "i-PRF", "A-PRF" and "L-PRF" with "microRNA", "exosomes", "extracellular vesicles", "mesenchymal stem cells", "osteogenic differentiation", "macrophage polarization" and "osseointegration", supplemented by targeted searches on exosome- and miRNA-functionalized titanium implants and on EV reporting standards. Priority was given to studies published from 2019 to 2026 that reported a mechanistic endpoint (gene or protein expression, miRNA profiling, EV characterization, stem-cell behaviour) and to human clinical studies reporting implant stability, radiographic bone or survival outcomes. Foundational descriptions of PRF and of osseointegration biology were retained regardless of date.

Each mechanistic finding was classified by model (in vitro, animal, human) and, where relevant, by tissue of origin, because a substantial proportion of the PRF-miRNA and PRF-EV literature comes from non-oral settings such as diabetic foot ulcers, cutaneous wounds, cartilage and endometrium. Such findings are presented as biologically plausible extrapolations, never as evidence of effect at the bone-implant interface. No predefined protocol, dual independent screening or formal risk-of-bias assessment was applied; the conclusions are therefore a critical synthesis rather than a pooled estimate of effect, consistent with the methodological scope of our previous review [4]. All bibliographic metadata were checked against PubMed records.

III. Osseointegration as a Signalling Problem: The Biological Window PRF Enters

The practical implication of the cell-communication model is that any adjunct placed in the osteotomy acts during a narrow temporal window, before the clot is replaced by granulation tissue and woven bone [1]. Two lines of implant research outside the PRF field show that this window is already regulated at the post-transcriptional and vesicular level. First, gene-activated titanium implants (surfaces loaded with plasmid DNA, miRNAs or small interfering RNAs through non-viral vectors) have enhanced osseointegration in animal models, particularly in compromised conditions such as osteoporosis, establishing nucleic-acid regulation as a legitimate therapeutic target at the bone-implant interface [9]. Second, implant surface topography changes the vesicular output of the host's own immune cells: macrophages cultured on 95-nm titania nanotubes released exosomes that enhanced the osteogenic gene expression of bone-marrow MSCs and the angiogenic capacity of endothelial cells, an effect traced by miRNA sequencing to exosomal miR-3473e acting through Akt1 [10].

Taken together, these findings reframe the peri-implant wound as an ongoing exchange of vesicles and miRNAs between macrophages, endothelial cells and MSCs. PRF does not create this conversation; it adds an autologous, platelet- and leukocyte-derived set of messages to it. The sections that follow examine each component of that message: what PRF does to MSCs, what miRNAs it carries or induces, what EVs it contains or can deliver, and how these converge on the macrophage as the integrating cell of osteoimmunology.

IV. Mesenchymal Stem Cells: Recruitment, Signalling and Dose

A. Recruitment and proliferation of local progenitors

The most directly implant-relevant MSC study used cells isolated from the Schneiderian membrane of rabbits. These cells expressed CD90 and CD105, lacked CD34 and CD45, and were capable of osteogenic, adipogenic and chondrogenic differentiation; PRF stimulated their proliferation, migration and osteogenic differentiation by up-regulating ERK1/2 signalling, and when mixed with deproteinized bovine bone in sinus floor elevation it increased the quantity and quality of new bone at 8 and 12 weeks [11]. This matters clinically because

the sinus membrane is the tissue lifted during sinus augmentation prior to or simultaneous with implant placement; the data suggest that PRF may act partly by activating a resident progenitor pool rather than by supplying osteogenic cells itself. Consistent with an MSC-supportive role, lyophilized PRF released growth factors that favoured bone-marrow MSC proliferation and osteogenic differentiation in vitro, and combined with osteogenic MSC sheet fragments it produced bone tissue in vivo without significant differences from fresh PRF [12].

B. Signalling pathways: TGF- β /Smad2/3, PDGF/Akt and BMP2/Smad1/5/8

Transcriptome-level evidence has begun to replace inference. RNA sequencing of dental pulp stem cells cultured in PRF-conditioned medium, compared with fetal bovine serum, identified 2,258 differentially expressed genes, with enrichment of TGF- β signalling and PDGF-binding pathways; phosphorylated Smad2/3 and Akt were increased, and pharmacological blockade of TGF- β (SB431542) or PDGF signalling (imatinib) reduced alkaline phosphatase activity and osteogenic marker expression [13]. In rat bone-marrow MSCs, PRF increased alkaline phosphatase activity and the expression of collagen I, BMP2, RUNX2 and osteocalcin, together with phosphorylated Smad1/5/8, placing PRF upstream of the canonical BMP2/Smad axis; a BMP activator produced stronger effects and an inhibitor attenuated them [14]. These pathways (ERK1/2, TGF- β /Smad2/3, PDGF/Akt and BMP2/Smad1/5/8) converge on RUNX2, the master transcription factor of osteoblast commitment.

C. Dose and lineage: more PRF is not necessarily better

Two observations complicate the simple narrative that PRF "drives" MSCs toward bone. In human gingival MSCs, 5% i-PRF increased proliferation at seven days, whereas 10% i-PRF reduced proliferation, lowered SPARC expression and did not induce spontaneous osteogenic differentiation (RUNX2 remained low); the authors concluded that excessive i-PRF concentrations can impair osteogenic induction [15]. In human adipose-derived stem cells, A-PRF extract increased both osteogenic (ALP, OPN, OCN, RUNX2) and adipogenic (PPAR γ 2, C/EBP α , FABP4) markers in a dose-dependent manner, together with paracrine (HIF-1 α , VEGF, IGF-2) and immunoregulatory genes [16]. PRF therefore appears to act as a broad, dose-sensitive amplifier of progenitor activity rather than as a lineage-specific osteoinductive signal; the osteogenic direction is supplied by the local context (bone matrix, implant surface chemistry and topography, mechanical loading). No study has yet defined a dose-response relationship for PRF volume or concentration at the bone-implant interface, which is a notable gap given how variably PRF is applied clinically.

V. MicroRNAs: The Post-Transcriptional Layer

A. PRF as a reservoir of stable miRNAs

The premise that PRF might transfer miRNAs to host cells requires, first, that miRNAs be present and stable in the clot. In canine PRF and other autologous blood products, miR-16, miR-21, miR-155 and miR-146a were abundant and remained stable under freezing at -30 °C for at least one month [17]. In human material, a recent review of PRF-derived EVs listed miR-21 and miR-146a among their regulatory cargo [18]. However, a comprehensive, protocol-stratified miRNA profile of human L-PRF, A-PRF and i-PRF, of the kind that would allow comparison between centrifugation protocols, has not been published, and species differences preclude direct transfer of the canine data.

B. miR-21 and the TGF- β /Smad axis in bone

miR-21 is the miRNA with the most consistent link to PRF-associated bone repair. In a rabbit calvarial model, alloxan-induced diabetes reduced miR-21 and MMP-9 and increased malondialdehyde, a lipid-peroxidation marker, at two weeks; grafting with L-PRF alone or combined with bovine hydroxyapatite improved miR-21 and MMP-9 and lowered malondialdehyde in diabetic animals, whereas the xenograft alone mainly increased superoxide dismutase activity [19]. In a 3D-bioprinted hydrogel scaffold loaded with i-PRF and nanosilicate (laponite), miRNA sequencing and functional validation showed that the combination promoted osteogenic differentiation of bone-marrow MSCs through miR-21- and miR-125a-mediated TGF- β /Smad signalling, while also favouring M2 macrophage polarization [20]. Read alongside the protein-level evidence that PRF activates Smad2/3 [13], these data allow a cautious working hypothesis: miR-21 may be one of the nodes that link PRF exposure to sustained TGF- β /Smad-dependent osteogenic commitment. This remains a hypothesis; the laponite scaffold study attributes the effect to synergy between ionic and growth-factor signals, not to PRF alone [20].

C. miR-155, hypoxia signalling and the compromised host

The strongest human miRNA data for PRF come from outside dentistry. In patients with diabetic foot ulcers, miR-155 was elevated in wound-margin tissue and correlated with Wagner grade; after 15 days of PRF, miR-155 decreased while HIF-1 α , VEGF and vascular density increased, and PRF was independently associated with faster healing (HR 3.21) [21]. The oral relevance is indirect but not trivial: diabetic patients are a population

in which delayed peri-implant healing is a recognized concern, and a PRF-induced shift in the miR-155/HIF-1 α /VEGF axis offers a plausible explanation for the angiogenic effect that clinicians attribute to PRF. It must be emphasized that this is an extrapolation from chronic cutaneous wounds to an acute osseous wound, and that no study has measured miR-155 in peri-implant tissue or crevicular fluid after PRF application.

VI. Extracellular Vesicles and Exosomes: From Cargo to Carrier

A. Terminology and the third-generation concept

A preliminary point of rigour is necessary. The MISEV2023 guidelines of the International Society for Extracellular Vesicles recommend the generic term "extracellular vesicle" unless a specific biogenesis pathway (such as endosomal origin for exosomes) has been demonstrated, and they set minimum requirements for separation, characterization and functional attribution [22]. Much of the dental literature uses "exosome" loosely for any small EV preparation, and this review therefore uses "EV" as the default term, reserving "exosome" for studies that used it. Within this framework, platelet-derived EVs have been proposed as essential mediators of platelet-concentrate-induced regeneration in periodontology and implant dentistry [23], and platelet-derived EVs of roughly 30-150 nm, carrying growth factors, miRNAs and lipids, have been positioned as a third generation of platelet concentrates, with a proposed sequential strategy of rapid PRP activation, PRF scaffolding and EV-mediated repair [8].

B. PRF-derived EVs: cargo and immunoregulatory activity

According to a recent review of PRF-derived EVs (PRF-EVs) appraised against MISEV2023 criteria, these vesicles, released during clot formation, carry TGF- β 1, VEGF and PDGF-A, matrix-regulating proteins (MMP-9, thrombospondin-1) and regulatory miRNAs (miR-21, miR-146a); they promote M2 macrophage polarization, suppress NF- κ B activation, rebalance the Th17/Treg axis and enhance fibroblast migration, angiogenesis and osteogenesis [18]. The same review noted that MSC-derived EVs have greater bioengineering potential, whereas PRF-EVs offer an advantage in biosafety and chairside feasibility, and that human trials remain limited in scale and duration [18]. For implant dentistry this suggests a division of roles that is worth testing: PRF as an autologous, low-cost EV source for routine cases, and engineered MSC-EVs for biologically compromised sites.

C. PRF and fibrin as EV carriers

Independently of its endogenous EV content, the fibrin matrix is an efficient delivery vehicle. Reviews of i-PRF and of drug-loaded platelet concentrates both identify exosomes among the biomolecules that PRF can retain and release gradually, and both stress that PRF's three-dimensional fibrin network gives it an advantage over PRP, whose burst-type release is short-lived, while also noting the absence of standardized loading protocols [3, 24]. Experimental data support this carrier function. A platelet-derived fibrin gel incorporated adipose MSC-EVs with high efficiency and released them continuously for up to four weeks, and the EV-loaded gel had stronger, synergic anti-inflammatory and matrix-remodelling effects than either component alone [25]. Combining menstrual-blood MSC-EVs with PRF in a rat full-thickness wound model accelerated closure by 23.2%, increased VEGF-A expression 1.9-fold and CD34-positive microvessel density 3.1-fold, and lowered IL-1 β and TNF- α while raising IL-10 and TGF- β [26]. In a rat endometrial-injury model, co-administration of i-PRF and MSC exosomes outperformed either treatment alone on fibrotic, inflammatory and angiogenic markers [27]. None of these models involves bone or titanium, but they consistently show that PRF plus MSC-EVs behaves as a combination rather than as two independent treatments.

D. EV-functionalized titanium: the missing link to PRF

A parallel line of implant research has immobilized EVs directly on titanium. Engineered exosomes enriched about eight-fold in untranslated Bmp2 mRNA, targeted to MSCs with an E7 peptide and anchored by a mussel-adhesive-protein coating, enhanced osteogenic differentiation of human MSCs and increased new bone formation and bone-implant contact in rat tibiae [28]. MSC exosomes bound to a zwitterionic choline-phosphate coating on 3D-printed porous titanium increased osteogenic gene expression and new bone formation in rabbit femoral defects at 4 and 12 weeks [29], and an E7- and exosome-functionalized hierarchical Ti6Al4V scaffold recruited and rejuvenated autologous MSCs and improved osseointegration in osteoporotic rats [30]. None of these studies used PRF. The convergence is nonetheless evident: if PRF can retain EVs and release them over weeks [25], a PRF coating could in principle serve as an autologous, chairside alternative to synthetic EV-immobilization chemistries. This remains untested, and the only randomized split-mouth trial of liquid-PRF-coated implants found no significant difference in stability, time to function, survival or marginal bone at one year [31], a reminder that a biologically attractive coating concept does not guarantee a clinical effect.

VII. Osteoimmunology: The Macrophage as Integrator

Across the MSC, miRNA and EV literature, the macrophage recurs as the cell through which PRF signals are translated into either regeneration or persistent inflammation. PRF-EVs [18], i-PRF-loaded scaffolds [20] and platelet-derived biomaterials more broadly have been reported to shift macrophages toward an M2 reparative phenotype, to promote regulatory T-cell differentiation in periodontitis [32] and, in osteoporotic bone, to influence the RANKL/OPG axis and restrain osteoclastogenesis [33]. Our earlier review summarized studies associating PRF with lower IL-1 β and TNF- α and higher IL-4 and IL-10 [4].

A 2026 transcriptomic study introduces an important counterweight. Bulk RNA sequencing of macrophage-like U937 cells exposed to PRF lysates revealed an inflammatory activation signature, with up-regulation of chemokines (including CCL2, CCL3, CCL4, CCL5 and CXCL8), IL1B, S100 alarmins and matrix metalloproteinases (MMP3, MMP9, MMP12), whereas THP-1 cells showed a more limited response centred on steroid biosynthesis [34]. The authors framed this early pro-inflammatory polarization as a physiological feature of wound healing rather than an adverse effect [34]. The two sets of findings are reconcilable if PRF is understood as a timing modulator: it may amplify the brief inflammatory signal required to launch healing and then, via EV cargo such as miR-146a and TGF- β 1, support resolution toward the M2 phase. This is consistent with the four-phase model of osseointegration, in which a controlled inflammatory phase is necessary and only its persistence is harmful [1]. It also implies that "anti-inflammatory" is too blunt a description of PRF, and that measurements taken at a single time point, or in a single cell line, can yield opposite conclusions.

VIII. From Mechanism to Clinic: What Human Implant Studies Show

The human evidence, summarized in Table 2, is modest and uses surrogate endpoints. A 2018 systematic review of 12 randomized trials found moderate evidence for a benefit of PRF on ridge preservation and in the early phase of osseointegration, with moderate or unclear risk of bias and heterogeneity that precluded meta-analysis [5]. A 2024 meta-analysis of PRF coating and secondary stability included 12 trials (456 implants), six at high and three at unclear risk of bias; pooled mean ISQ differences of roughly 2.5 to 3.4 favoured PRF at four, six and at least 12 weeks, but the certainty of evidence was low [6]. The 2026 scoping review of 19 human studies found that resonance frequency analysis (ISQ) was the most common outcome and frequently suggested a transient improvement in secondary stability during the first weeks, while studies with 6-12 months of follow-up did not establish consistent superiority in marginal bone preservation or survival; relative centrifugal force, rotor geometry and tube composition were commonly unreported [7].

Individual trials illustrate the spread. In a split-mouth study of 20 patients, sockets irrigated with i-PRF showed a rise in ISQ from 68.9 to 74.2 over three months versus 73.3 to 73.7 in controls, although the test implants started from a lower baseline [35]. A 40-patient comparative study reported a mean ISQ of 88.6 with PRF versus 72.8 without it at six months [36], an unusually large difference that warrants independent replication. In 30 immediate implants in the aesthetic zone, PRF did not change intergroup marginal bone levels, although intragroup bone density increased, an effect the authors described as promising but transitory [37]. The double-blinded split-mouth RCT of liquid-PRF coating found no significant difference in any parameter at one year [31]. Crucially, none of these clinical studies measured a molecular or cellular endpoint (miRNA expression, EV content, MSC mobilization or macrophage phenotype), so the mechanisms discussed in Sections IV-VII cannot yet be linked to the clinical outcomes they are invoked to explain.

IX. An Integrated Model and the Release-Window Hypothesis

Table 1 organizes the evidence into four mechanistic axes and grades each by model. Placed on the timeline of osseointegration [1], a coherent sequence emerges. In the first hours to days, the PRF fibrin network, platelets, leukocytes and EVs join the native clot, and macrophages mount a transient inflammatory response that PRF may intensify [34]. Over the following one to two weeks, growth factors and EV cargo activate ERK1/2, TGF- β /Smad2/3, PDGF/Akt and BMP2/Smad1/5/8 signalling in local MSCs [11, 13, 14], while miRNAs such as miR-21 appear to help sustain osteogenic commitment [19, 20] and immunoregulatory cargo supports the shift toward M2 macrophages [18]. Remodelling and the establishment of load-bearing bone, which occur over months, lie largely outside the period during which PRF actively releases its bioactive content, which for the fibrin matrix has been described as extending to approximately 14 days [4].

We propose, as a testable interpretation rather than a conclusion, a "release-window hypothesis": because PRF acts within a short biological window while clinical osseointegration is judged over months, any durable effect would have to be carried forward by lasting changes in the behaviour of the cells PRF reaches (for example, miRNA-mediated reprogramming of MSC and macrophage phenotypes). Where that reprogramming does not occur, a transient early advantage in secondary stability would be expected to dissipate, which is precisely the pattern reported in the clinical literature [7, 37]. The hypothesis generates specific predictions: early ISQ gains should correlate with early changes in crevicular-fluid miRNAs or EV markers; sustained EV release from

engineered PRF-EV constructs should prolong the effect; and biologically compromised hosts, in whom the native signal is weaker, should show larger relative benefits.

Table 1. Mechanistic axes of PRF action relevant to osseointegration, with model and evidence tier

Axis	Key mediators / findings	Model (tissue)	Evidence tier and relevance
MSC recruitment and commitment	ERK1/2 activation; proliferation, migration and osteogenesis of sinus-membrane MSCs; more new bone with xenograft [11]. TGF- β /Smad2/3 and PDGF/Akt [13]; BMP2/Smad1/5/8, RUNX2, OCN [14]	In vitro + rabbit sinus; human DPSCs; rat BMSCs	Preclinical; directly relevant to sinus augmentation and osteotomy healing
Dose dependency	5% i-PRF raised, 10% lowered gingival MSC proliferation; no osteoinduction at high dose [15]; A-PRF extract raised osteogenic and adipogenic markers [16]	Human gingival MSCs; human ASCs (in vitro)	In vitro; PRF acts as an amplifier, not lineage-specific; no clinical dose-response defined
MicroRNAs	miR-16, -21, -146a, -155 stable in PRF [17]; L-PRF restored miR-21 in diabetic bone [19]; miR-21/-125a drive TGF- β /Smad with i-PRF scaffold [20]; PRF lowered miR-155, raised HIF-1 α /VEGF [21]	Canine blood; diabetic rabbit calvaria; rat cranial defect; human diabetic foot ulcer	Preclinical in bone; human data only in non-oral wounds; no peri-implant miRNA study
Extracellular vesicles	PRF-EVs carry TGF- β 1, VEGF, PDGF-A, miR-21, miR-146a; M2 shift [18]; fibrin gel releases MSC-EVs up to 4 weeks [25]; EV-coated titanium increases bone-implant contact [28-30]	Review; in vitro; rat, rabbit and osteoporotic rat implants (without PRF)	Preclinical; PRF-EV implant coatings untested; EV reporting per MISEV2023 [22] often incomplete
Osteoimmunology	M2 polarization, Treg support, RANKL/OPG modulation [32, 33]; early inflammatory transcriptome in U937 macrophages [34]	Reviews; human cell lines	Mixed and time-dependent; PRF best viewed as a timing modulator

ASC, adipose-derived stem cell; BMSC, bone-marrow mesenchymal stem cell; DPSC, dental pulp stem cell; EV, extracellular vesicle; MSC, mesenchymal stem/stromal cell.

Table 2. Selected human evidence on PRF and dental implant osseointegration

Study	Design / sample	Main finding	Mechanistic endpoint measured?
Strauss et al., 2018 [5]	Systematic review, 12 RCTs	Moderate evidence for ridge preservation and early osseointegration phase	No
Khatti et al., 2024 [6]	SR/meta-analysis, 12 trials, 456 implants (secondary stability)	ISQ MD about 2.5-3.4 favouring PRF; 9 of 12 trials high/unclear risk of bias; low certainty	No
Gitanjali et al., 2026 [7]	Scoping review, 19 human studies	Transient early ISQ gains; no durable superiority in bone level or survival; RCF often unreported	No
Fernandes et al., 2022 [31]	Double-blind split-mouth RCT, 15 patients / 30 implants, 1 year	Liquid-PRF coating: no significant difference in ISQ, function time, survival or bone loss	Cytokine profile of liquid PRF only
Hasanzade et al., 2025 [35]	Split-mouth, 20 patients, 3 months	Greater ISQ gain with i-PRF irrigation; lower baseline in test group	No
Jalaluddin et al., 2025 [36]	Comparative, 40 patients, 6 months	ISQ 88.6 vs 72.8 favouring PRF; requires replication	No
Iram et al., 2024 [37]	Prospective, 30 immediate implants	No intergroup marginal bone difference; intragroup density gain, "transitory"	No

ISQ, implant stability quotient; MD, mean difference; RCF, relative centrifugal force; RCT, randomized controlled trial; SR, systematic review.

X. Discussion: Limitations and Research Agenda

The central finding of this review is a mismatch in evidence levels. The mechanistic model is increasingly coherent (PRF acts on MSCs through identifiable kinase and Smad pathways, carries stable miRNAs and EVs, and modulates macrophage behaviour), but almost every link in that chain has been demonstrated in vitro, in animals, or in non-oral human tissues. Several limitations follow. First, extrapolation across tissues and species is extensive: the strongest human miRNA data come from diabetic foot ulcers [21], the miRNA stability data from dogs [17], and the PRF-plus-EV synergy data from skin, cartilage and endometrium [25-27]. Second, EV terminology and characterization are inconsistent, and few dental studies meet MISEV2023 reporting standards [22]. Third, PRF itself is not a single product: centrifugation force, time, rotor and tube type alter its composition, and these parameters are frequently unreported [4, 7], which makes it impossible to know whether two "PRF" studies tested the same material. Fourth, dose matters in both directions [15, 16], yet clinical protocols rarely define it. Fifth, the clinical literature relies on surrogate endpoints (principally ISQ) over short follow-up and has never included a molecular endpoint.

A focused research agenda can be derived from these gaps. (1) Characterize the miRNA and EV content of human L-PRF, A-PRF and i-PRF under explicitly reported centrifugation protocols, following MISEV2023. (2) Embed mechanistic endpoints in clinical trials, for example serial peri-implant crevicular fluid sampling for miR-21, miR-146a and miR-155 and EV markers during the first eight weeks, so that early ISQ changes can be related to biology. (3) Establish dose-response relationships for PRF volume and concentration at the osteotomy. (4) Test PRF-based EV retention as an autologous implant-surface strategy in preclinical implant models, benchmarked against synthetic EV-immobilization coatings [28, 29], with priority for compromised hosts such as patients with diabetes or osteoporosis, in whom the relative benefit is predicted to be greatest [19, 33]. (5) Distinguish regulatory and cost pathways: minimally manipulated chairside PRF differs fundamentally from isolated or engineered EV products, and the low cost and simplicity of PRF, which favour its adoption in public and lower-cost services [4], would be lost if its benefits could only be reproduced with laboratory-manufactured vesicles.

This review has its own limitations. It is narrative rather than systematic, it relied on a single database, and the grading of evidence tiers reflects the author's judgement rather than a formal instrument. The integrated model and the release-window hypothesis are interpretive constructs intended to generate testable predictions, not established mechanisms.

XI. Conclusion

PRF's contribution to dental implant osseointegration is better explained by a multilayered signalling model than by growth-factor release alone. Preclinical evidence indicates that PRF activates local mesenchymal stem cells through ERK1/2, TGF- β /Smad2/3, PDGF/Akt and BMP2/Smad1/5/8 signalling in a dose-dependent way, carries stable microRNAs such as miR-21 and miR-146a, contains and can deliver extracellular vesicles, and modulates macrophage behaviour in a time-dependent manner. Human implant studies, however, show mainly transient early gains in stability and have not measured any of these mechanisms. Until molecular endpoints are incorporated into standardized clinical trials, microRNAs, exosomes and mesenchymal stem cells should be regarded as a plausible and increasingly well-supported biological rationale for PRF, not yet as a validated explanation of its clinical effect.

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