Japanese Dental Science Review 58 (2022) 89–123

Contents lists available at ScienceDirect

Japanese Dental Science Review

journal homepage: www.elsevier.com/locate/jdsr

Review article

The application of injectable platelet-rich fibrin in regenerative dentistry: A systematic scoping review of *In vitro* and *In vivo* studies☆

Nima Farshidfar (DDS)[a,b,*,1], Dana Jafarpour (DDS)[c,1], Parsa Firoozi (DDS)[d,e], Sarina Sahmeddini (DDS)[b], Shahram Hamedani (DDS, MSc)[f], Raphael Freitas de Souza (DDS, MSc, PhD)[c], Lobat Tayebi (PhD)[g,**]

[a] *Orthodontic Research Center, School of Dentistry, Shiraz University of Medical Sciences, Shiraz, Iran*
[b] *Student Research Committee, Shiraz University of Medical Sciences, Shiraz, Iran*
[c] *Faculty of Dental Medicine and Oral Health Sciences, McGill University, Montreal, QC, Canada*
[d] *Student Research Committee, School of Dentistry, Zanjan University of Medical Sciences, Zanjan, Iran*
[e] *Department of Oral and Maxillofacial Surgery, School of Dentistry, Zanjan University of Medical Sciences, Zanjan, Iran*
[f] *Oral and Dental Disease Research Center, School of Dentistry, Shiraz University of Medical Sciences,Shiraz, Iran*
[g] *Marquette University School of Dentistry, Milwaukee, WI, USA*

A R T I C L E I N F O

Article history:
Received 7 August 2021
Received in revised form 24 January 2022
Accepted 20 February 2022

Keywords:
Platelet-rich Fibrin
I-PRF
Biomaterial
Tissue Regeneration
Regenerative Dentistry

A B S T R A C T

Background: Ongoing research in the dental field has begun to focus on the use of injectable platelet-rich fibrin (I-PRF) as a regenerative tool with the potential to prompt tissue regeneration. In this regard, this systematic scoping review aimed to collect, map, and appraise the *in vitro* and *in vivo* studies regarding the role of I-PRF in or soft and hard tissue regeneration in relation to oral and maxillofacial structures.
Methods: A systematic electronic search of *Medline, Scopus, Web of Science, and Embase* databases was performed from 2000 to December 2021 using a combination of keywords. All *in vitro* and *in vivo* studies, written in English and concerning the potential role of I-PRF in regenerative dentistry were considered.
Results: In total, 18 *in vitro* studies, 5 animal studies, 6 case reports, and 31 clinical studies have evaluated the effect of I-PRF on oral and maxillofacial soft and hard tissue regeneration. The investigated studies verified the anti-inflammatory, anti-microbial efficacy and the positive effects of I-PRF application for wound, periodontal, bone, cartilage, and pulp regeneration, as well as acceleration in tooth movement during orthodontic treatment.
Conclusions: Current literature approves the feasibility of I-PRF application as a promising regenerative adjunct to dental procedures.

Contents

☆ Scientific Field of Dental Science: Dental Biomaterials.

* Correspondence to: School of Dentistry, Shiraz University of Medical Sciences, Ghom Abad Street, Shiraz 71348-14336, Iran.
** Correspondence to: Marquette University School of Dentistry, Milwaukee, WI 53233, USA.
E-mail addresses: n.farshidfar@icloud.com (N. Farshidfar), dana.jafarpour@mail.mcgill.ca (D. Jafarpour), parsafir2@gmail.com (P. Firoozi), sarinasahmeddini@yahoo.com (S. Sahmeddini), shahramhamedani@yahoo.com (S. Hamedani), raphael.desouza@mcgill.ca (R.F. de Souza), lobat.tayebi@marquette.edu (L. Tayebi).
[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.jdsr.2022.02.003

1. Introduction

One of the greatest challenges that researchers are facing today is producing a biomaterial, which can be employed to enhance tissue regeneration with maximum predictability [1]. Though our knowledge on tissue healing process is still insufficient, it is clear that platelets can play a significant role in the tissue regenerative procedures [2].

The mechanisms of platelets in regeneration were demonstrated in the 1970s [3]. Platelets produce growth factors in their alpha-granules, which are accountable for cell division, differentiation, induction, and migration, and for neovascularization and collagen synthesis. Hence, they are known as potentiated cells for regeneration which can facilitate tissue regeneration [4].

Various platelet-rich concentrates, such as platelet-rich plasma (PRP) and platelet-rich fibrin (PRF) have been proposed and utilized for tissue regeneration in several *in vitro* and *in vivo* studies [3]. Nevertheless, PRF has many benefits over PRP, including easy handling, low cost, and the lack of anticoagulant or bovine thrombin, which reduces biochemical alteration and risks associated with the use of bovine thrombin [5]. For about three decades, PRF has been used for regenerative purposes in dentistry [6]. Additionally, PRF has the potential to be used in fields other than dentistry, such as maxillofacial surgery and orthopedic surgery [7–9].

In 2014, by adjusting spin centrifugation forces, injectable platelet-rich fibrin (I-PRF) was developed. The blood centrifuged in non-glass centrifugation tubes at lower centrifugation speeds resulted in a flowable PRF called I-PRF [10]. I-PRF is a newly formed platelet concentrate enriched with leukocytes which can promote both soft and hard tissue regeneration phenomena [10,11]. Since I-PRF remains liquid for roughly 15 min, it will provide dental practitioners with a further practical form of PRF [12]. Following application, the human liquid fibrinogen in I-PRF is gradually transformed to a growth factor-rich PRF clot, which releases continuously over 10–14 days [13].

Up to now, several *in vitro* and *in vivo* studies have been carried out concerning the role of I-PRF in the enhancement of wound healing, the acceleration of orthodontic tooth movement, and the regeneration of bone, periodontal, cartilage, and pulp tissues [14–21]. On this basis, I-PRF is able to enhance the potential of intrinsic tissue regeneration by inducing human mesenchymal stem cells (MSCs) proliferation and migration, and by triggering osteogenic differentiation of MSCs [21,22]. I-PRF has also been reported to have greater anti-inflammatory and anti-microbial activity against many pathogens, which can contribute to faster tissue regeneration [23,24]. On the other hand, I-PRF is commonly used in regenerative dentistry as an injectable biomaterial, as a carrier for various biomolecules, or in conjunction with other biomaterials for a variety of clinical applications. Clinicians have recently used this method to facilitate the agglomeration or coating of biomaterials in order to improve the healing process of both soft and hard tissues [25–27].

Given the aforementioned advantages of utilizing I-PRF and since there is a lack of a comprehensive review regarding the application

Table 1
Eligibility criteria for the present systematic scoping review.

Domain	Inclusion criteria	Exclusion criteria
Participants	– Cells, microorganisms, and tissues related to dental, oral and maxillofacial structures	– Dermal cells and tissues
Intervention	– Using liquid- or injectable-PRF developed by low-speed centrifugation concept for cell proliferation, migration, viability, morphology, mineralization, differentiation, preventing microorganism growth, healing damaged tissues, and drug delivery in dental, oral and maxillofacial structures	– Using other kinds of platelet concentrates only such as PRGF, CGF, PRP, PRF, L-PRF, A-PRF, C-PRF, Alb-PRF, PRF exudates, PRF lysate
Comparison	– No treatment or receiving other treatments	–
Outcome	– Cell and microorganism behavior, and tissue response after treatment	–
Study Design	– In-vitro studies, ex-vivo studies, animal studies, non-comparative studies, case reports, case series and prospective/retrospective clinical trials	– Narrative reviews, systematic reviews with or without meta-analysis, letters to the editors, short communications

of I-PRF in regenerative dentistry, this systematic scoping review aimed to map the current literature and to review the articles published to date concerning the potential role of I-PRF in regenerative dentistry where this injectable autologous biomaterial is used to promote soft and hard tissue regeneration.

2. Material and methods

2.1. Development of a protocol

The protocol followed in this study was adopted from the guidelines of the Joanna Briggs Institute on systematic scoping reviews [28]. A research question comprising the inclusion criteria for the participants, intervention, comparison, outcome, and study design (PICOS) was defined prior to starting the review, which is shown in Table 1.

2.2. Information sources and search strategy

A systematic electronic search of four databases (*Medline, Scopus, Web of Science, and Embase*) was performed, which is illustrated in Table 2. Articles published from 2000 up to December 1, 2021 were considered. An extra hand search was performed on bibliographies of retrieved papers as well as other related published systematic and narrative reviews for possible inclusion in the study.

2.3. Eligibility criteria

Only peer-reviewed, published articles pertaining to our PICOS question were considered in this review (Table 1). Studies in other languages other than English were excluded from our review considering the linguistic competency of the research team.

2.4. Selection of studies

Records retrieved by the systematic search were independently screened by four reviewers (NF, DJ, and PF, SS) according to the inclusion and exclusion criteria (Fig. 1). After initial screening through titles and abstracts, the full texts of the retrieved abstracts were independently screened by the reviewers. Disagreements were resolved by discussion among the research team (SH, RFS, and LT). Finally, articles, which met the inclusion criteria and linguistic capacities of the research team, were included for data extraction and analysis.

2.5. Data extraction

Data extraction was carried out based on the inclusion criteria. A custom-made data collection form was established to collect information from the final selection of studies. Data including the authors name, year of publication, type of studies, characteristics of cells/drugs/animals/humans, preparation method of I-PRF, and details of the interventions, methods, and outcome measures were compiled in Tables 3–6.

3. Results and discussion

3.1. Selection of studies

In total, the initial search strategies generated 837 articles. After duplicate removal, 509 abstracts remained for title and abstract evaluation. A total of 431 papers were excluded due to mismatch with our search criteria and 78 articles were retained for final full text review. Finally, 60 articles including 18 *in vitro* studies, 5 animal studies, 6 case reports, and 31 clinical studies, which have so far

Table 2
Strategies for database searches, from 2000 up to December 1, 2021.

Database	Search strategy	Hits
Medline	((((((((((Injectable platelet rich fibrin[Title/Abstract]) OR (liquid platelet rich fibrin[Title/Abstract])) OR (Flowable platelet rich fibrin[Title/Abstract])) OR (i-PRF[Title/Abstract])) OR (injectable[Title/Abstract] AND platelet rich fibrin[Title/Abstract])) OR (liquid[Title/Abstract] AND platelet rich fibrin[Title/Abstract])) OR (Flowable[Title/Abstract] AND platelet rich fibrin[Title/Abstract])) OR (Injectable[Title/Abstract] AND PRF[Title/Abstract])) OR (liquid[Title/Abstract] AND PRF[Title/Abstract])) OR (Flowable[Title/Abstract] AND PRF[Title/Abstract])	167
Scopus	(TITLE-ABS-KEY ("Injectable platelet rich fibrin") OR TITLE-ABS-KEY ("liquid platelet rich fibrin") OR TITLE-ABS-KEY ("Flowable platelet rich fibrin") OR TITLE-ABS-KEY ("i-PRF") OR TITLE-ABS-KEY ("injectable" AND "platelet rich fibrin") OR TITLE-ABS-KEY ("liquid" AND "platelet rich fibrin") OR TITLE-ABS-KEY ("Flowable" AND "platelet rich fibrin") OR TITLE-ABS-KEY ("Injectable" AND "PRF") OR TITLE-ABS-KEY ("liquid" AND "PRF") OR TITLE-ABS-KEY ("Flowable" AND "PRF")) AND PUBYEAR > 2000	237
Web of Science	TOPIC: (Injectable platelet rich fibrin) OR TOPIC: (liquid platelet rich fibrin) OR TOPIC: (Flowable platelet rich fibrin) OR TOPIC: (i-PRF) OR TOPIC: (injectable AND platelet rich fibrin) OR TOPIC: (liquid AND platelet rich fibrin) OR TOPIC: (Flowable AND platelet rich fibrin) OR TOPIC: (Injectable AND PRF) OR TOPIC: (liquid AND PRF) OR TOPIC: (Flowable AND PRF)	251
Embase	('injectable platelet rich fibrin':ab,ti OR 'liquid platelet rich fibrin':ab,ti OR 'flowable platelet rich fibrin':ab,ti OR 'i prf':ab,ti OR (injectable:ab,ti AND 'platelet rich fibrin':ab,ti) OR (liquid:ab,ti AND 'platelet rich fibrin':ab,ti) OR (flowable:ab,ti AND 'platelet rich fibrin':ab,ti) OR (injectable:ab,ti AND prf:ab,ti) OR (liquid:ab,ti AND prf:ab,ti) OR (flowable:ab,ti AND prf:ab,ti)) AND [2000–2021]/py	182

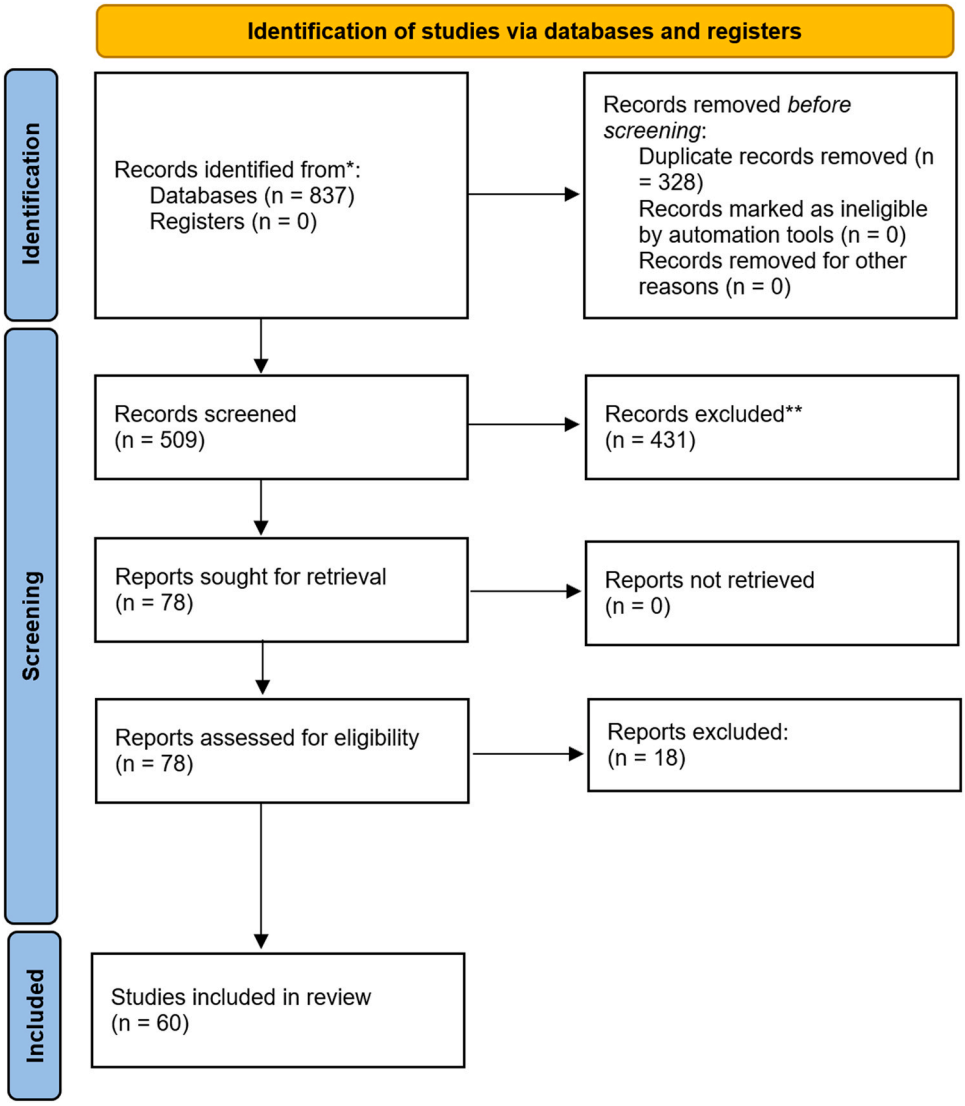

Fig. 1. : PRISMA 2020 flow chart.

evaluated the effect of I-PRF on soft and hard tissue regeneration related to oral and maxillofacial structures, were selected. The selected studies were classified under the following subheadings: wound healing and anti-inflammatory efficacy (n = 9), anti-microbial efficacy (n = 4), periodontal regeneration (n = 13), bone regeneration (n = 22), cartilage regeneration (n = 7), orthodontic tooth movement (n = 5), and pulp regeneration and drug delivery (n = 3). Studies by Mu et al. [29], Dohle et al. [30], and Rafiee et al. [31] touched upon two regenerative potentials of I-PRF; therefore, they were included in two categories. The Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) Flow Diagram 2020 in Fig. 1 depicts the flow of included studies through each phase of the review process. Additionally, Fig. 2 illustrates a progressive increase in the frequency of publications from 2017 to 2021.

3.2. I-PRF preparation method

The I-PRF preparation method differs according to the centrifugation time and speed, centrifuge device, and the site from which sample is collected [32].

3.2.1. Centrifugation time and speed

Most of the included studies used an optimum centrifugation speed of 700 rate per minute (rpm) for obtaining I-PRF, based on the notion that the number of platelets, inflammatory factors, and cytokine significantly increases with a reduction in relative centrifugal force (RCF) [10,33].

In studies evaluating the wound healing, anti-inflammatory, and anti-microbial efficacy of I-PRF, the process of I-PRF preparation was almost the same, in that blood collected in tubes devoid of anticoagulant was centrifuged at a speed of 700 rpm for three minutes with a centrifugal device. Only Kiziltoprak et al. [34] used a protocol of 2300 rpm for three minutes, and Jasmine et al. [35] implemented a method of 1000 rpm for five minutes. The method of I-PRF preparation was mostly similar for bone regeneration purposes in which I-PRF was attained by centrifuging the tubes in a centrifuge at 700 rpm for three minutes. The exceptions were the study by Valladão et al. [36] in which I-PRF was acquired after centrifuging two non-ridged tubes holding 8 ml blood at 2700 rpm for three minutes in a centrifugal machine, the research by Kyyak et al. [37] which centrifuged the tubes at 1200 rpm for eight minutes, Irdem et al.

Table 3
A summary of the included *in-vitro* studies regarding in the application of I-PRF in regenerative dentistry.

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
Jasmine et al.[35] (2020)	Anti-microbial Efficacy	To evaluate the anti-microbial and anti-biofilm activity of I-PRF against pathogenic oral staphylococcus isolates.	- Staphylococcus aureus (isolated from patients with oral and dental abscess) - Staphylococcus epidermis (isolated from patients with oral and dental abscess) - Staphylococcus epidermis ATCC 35,984 (Weak biofilm producer) - Staphylococcus epidermis ATCC 12,228 (non-biofilm producer)	1000 rpm 5 min 37 °C (NR)	Upper layer	**Anti-microbial Activity:** G1) Staphylococcus epidermis ATCC 12,228 + I-PRF G2) Staphylococcus epidermis ATCC 35,984 + I-PRF G3) Staphylococcus epidermis + I-PRF G4) Staphylococcus aureus + I-PRF **Anti-biofilm Activity:** G1) non-biofilm producer (Staphylococcus epidermis ATCC 12,228) + I-PRF G2) Weak biofilm producer (Staphylococcus epidermis ATCC 35,984) + I-PRF G3) Moderate biofilm producer (Staphylococcus epidermis) + I-PRF G4) Strong biofilm producer (Staphylococcus aureus) + I-PRF	**Anti-microbial Assay by Broth Microdilution Method:** MIC of I-PRF for non-biofilm producing bacteria→ 80 ml/ml MIC of I-PRF for weak, moderate and strong producing bacteria→ 160 ml/ml MBC of I-PRF for non-biofilm producing bacteria → 160 ml/ml MBC of I-PRF for weak, moderate and strong producing bacteria → 240 ml/ml **Live/Dead Microbial Assay:** I-PRF at MIC → > 50% bacterial cells necrosis I-PRF at MBC → 100% bacterial cells necrosis **Biofilm Inhibitory Assay by Semi-quantitative Plate Method:** I-PRF at MIC → Significant reduction ($P < 0.05$) against weak, moderate and strong biofilm producers I-PRF at MBC → All biofilm producers were unable to produce the biofilm.
Kour et al.[56] (2018)	Anti-microbial Efficacy	To evaluate the anti-microbial effect of I-PRF against two periodontal pathogens compared with PRP and PRF.	- Porphyromonas gingivalis - Aggregatibacter actinomycetemcomitans	700 rpm 3 min - (NR)	NR	G1) Porphyromonas gingivalis + PRP/PRF/I-PRF G2) Aggregatibacter actinomycetemcomitans + PRP/PRF/I-PRF	**Anti-microbial Assay by Disc Diffusion Method:** G1 → I-PRF > PRP and PRF ($P < 0.05$) G2 → PRP > PRF and I-PRF ($P < 0.05$)
Karde et al.[24] (2017)	Anti-microbial Efficacy	To evaluate the anti-microbial effect of I-PRF compared with PRP, PRF and whole blood.	- Supragingival plaque samples of individuals who volunteered for blood sample.	700 rpm 3–4 min - (NR)	NR	G1) Supragingival plaque samples + PRP/PRF/I-PRF/Whole blood	**Anti-bacterial Assay by Disc Diffusion Method:** G1 → I-PRF > PRF > PRP ($P > 0.05$)
Kyyak et al.[37] (2021)	Bone Regeneration	To compare the effect of XBSM with and without I-PRF for viability and metabolic activity of human osteoblasts as well as expression of ALP, BMP-2, and OCN.	- Human osteoblasts	1200 rpm 8 min - (Duo Centrifuge, Carasco, Italy)	NR	G1) XBSM G2) XBSM + I-PRF	**Human Osteoblasts Viability Assay and Metabolic Acivity by MTT Assay (On 3rd,7th, and 10th Day):** Day 3 → Increased viability, and metabolic activity (G2 > G1) Day 7 → The highest viability and metabolic activity (G2 > G1) Day 10 → Viability and metabolic activity tend to decline. **Osteogenic-related Gene Expression by RT-qPCR (On 3rd,7th and 10th Day):** Day 3 → No changes in ALP, and increased expression of OCN & BMP-2 (G2 > G1) Day 7 and 10 → No changes in ALP, and BMP-2, and increased expression of OCN (G2 > G1)
Murdiastuti et al.[65](2021)	Bone Regeneration	To evaluate the effect of I-PRF and freeze-dried	- Human osteoblasts	700 rpm 3 min Room	Upper layer	G1) Control G2) I-PRF G3) Freeze-Dried Homologous	**Histological Staining Analysis:** **1) H&E Staining (On 7th Day):**

(*continued on next page*)

Table 3 (*continued*)

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
		homologous PRP on human osteoblasts (osteogenesis).		temprature (NR)		PRP (20 KGy) G4) Freeze-Dried Homologous PRP (25 KGy)	G2 had the highest number of osteocytes.
Shah et al.[66] (2021)	Bone Regeneration	To assess the response of osteoblast-like cell line (MG-63) coating of I-PRF on titanium disks.	- MG-63 osteoblastic cells	700 rpm 3min - (IntraSpin, Florida, USA)	Orange layer	G1) Titanium Disks (n = 5) G2) Titanium Disks Coated with I-PRF (n = 5)	**MG-63 Osteoblastic Cells Proliferation by CCK-8 Assay (On the 1st, 7th, 14th, and 21st Day):** G2 > G1 (P < 0.001) **MG-63 Osteoblastic Cells Mineralization by Alizarin Red Staining (On 21st Day):** G2 > G1 (P < 0.001) **ALP Activity (On the 1st, 7th, 14th, 21st Day):** G2 > G1 (P < 0.001)
Kyyak et al.[13] (2020)	Bone Regeneration	To evaluate the effect of an ABSM and a XBSM with and without I-PRF on cell characteristics of human osteoblasts.	- Human osteoblasts	700 rpm 3 min Room temperature (Duo Centrifuge, Cologne, Germany)	Upper layer	G1) ABSM G2) ABSM + I-PRF G3) XBSM G4) XBSM + I-PRF	**Human Osteoblasts Viability Assay, Migration by Scratch Assay, and Proliferation by MTT Assay (On 3rd, 7th and 10th Day):** Day 3 → Increased viability, migration and proliferation for G2 Day 7 → Increased viability and proliferation (G2/G4 > G1/G3) Day 10 → Increased viability, proliferation and migration (G2/G4 > G1/G3) **Osteogenic-related Gene Expression by RT-qPCR (On 3rd, 7th and 10th Day):** Days 3 and 7 → Highest expression of alkaline phosphatase for G2/G4, highest BMP-2 expression for G2, increased expression of OCN for G2/G4, and lowest expression for G1. Day 10 → Increased expression of alkaline phosphatase for G1/G2, and increased expression of OCN for G2.
Fernández-Medina et al. [64] (2019)	Bone Regeneration	To investigate the composition and bioactivity of four common clinical-grade hemoderivates prepared using standardized methods.	- Human osteoblasts	700 rpm 3 min - (NR)	Orange supernatant layer	G1) Control G2) Clot (20%, 40%, 60%, 80% and 100% v/v) G3) A-PRF (20%, 40%, 60%, 80% and 100% v/v) G4) I-PRF (20%, 40%, 60%, 80% and 100% v/v) G5) P-PRP (20%, 40%, 60%, 80% and 100% v/v) G6) L-PRP (20%, 40%, 60%, 80% and 100% v/v)	**Growth Factor Release of IGF-I, VEGF, PDGF-BB, and BMP-2 by ELISA (On 1st, 3rd, 7th and 14th Days):** Early time points → A significant cumulative release of IGF-I and PDGF-BB for G3/G6, and a significant cumulative release of BMP-2 and VEGF for all groups. **Human Osteoblasts Viability by Live/Dead Assay (At 24 and 72 h), Metabolic Activity by Alamar Blue Assay (At 24 and 72 h), and Migration by Scratch Assay (At 6 and 24 h):** Cell viability, metabolic activity and migration assay → Detrimental effect when the concentration of all groups was ≥ 60%

(*continued on next page*)

Table 3 (*continued*)

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
							Human Osteoblast Mineralization by Alizarin Red Staining (On 14th, and 21st Day): Day 21 → Superior mineralization properties for G4 compared to all groups. Days 14 and 21 → A negative impact of A-PRF was demonstrated at high concentrations.
Wang et al.[63] (2018)	Bone Regeneration	To investigate the effect of I-PRF on osteoblast behavior compared to traditional PRP.	- Human Osteoblasts	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Upper layer	G1) Control G2) PRP G3) I-PRF	**Human Osteoblasts Viability by Live/Dead Assay (At 24 h):** Viability → High survival rates of cells for all groups. **Human Osteoblasts Migration Assay (At 24 h), Adhesion by DAPI Staining (At 2, 4 and 8 h), and Proliferation by CCK-8 Assay (On 1st, 3rd and 5th Day):** Migration → G3 > G2 > G1 ($P < 0.05$) Adhesion → No difference between all groups. Proliferation → G3 = G2 > G1 on 3rd and G3 > G2 > G1 on 5th day ($P < 0.05$) **Human Osteoblast Mineralization by ALP Assay (On 7th Day) and Alizarin Red Staining (On 14th Day):** G3 > G2 > G1 ($P < 0.05$) **Osteogenic-related Gene Expression by RT-qPCR on (3rd and 14th Day) and Immunohistochemical Staining of Osteocalcin (on 14th Day):** Expression of ALP on 3rd day → G3 > G2 = G1 Expression of ALP, RUNX2 and OCN on 14th day → G3 > G2 > G1 ($P < 0.05$) Expression of COL1 on 14th day → G3 = G2 > G1
Dohle et al.[30] (2018)	Bone Regeneration, Wound Healing and Anti-inflammatory Efficacy	To examine that the addition of I-PRF would result in an induction of wound healing processes and might positively influence the process of angiogenesis *via* inflammatory processes in the co-culture.	- Human OECs - Human pOBs	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	G1) OEC G2) pOB G3) co-culture of OEC + pOB G4) I-PRF G5) I-PRF + OEC G6) I-PRF + pOB G7) I-PRF + co-culture of OEC + pOB	**Histological Staining Analysis: 1) H&E, and 2) Immunohistochemical Staining of CD31, CD68, CD45 and Osteopontin (At 24 h and on 7th Day):** 7th day → Formation of lumina and microvessel-like structures in the I-PRF/co-culture complexes. **Protein Quantification by ELISA, and Gene Expression by RT-qPCR (At 24 h and on 7th Day):** I-PRF + Co-culture of OEC and pOB → Angiogenic activation of OECs + Upregulation of wound healing-associated factors (PDGF-BB, ICAM-1, and E-selectin) + Higher expression of the proangiogenic factor

(*continued on next page*)

Table 3 (*continued*)

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
Zheng et al.[58] (2020)	Periodontal Regeneration	To evaluate the biological effect of PRP and I-PRF on hPDLCs *in vitro*.	- hPDLCs	700 rpm 3 min - (IntraSpin, Florida, USA)	Upper layer	G1) hPDLCs G2) hPDLCs + PRP G3) hPDLCs + I-PRF **Inflammatory Environment:** G1) hPDLCs G2) hPDLCs + LPS G3) hPDLCs + I-PRF G4) hPDLCs + LPS + I-PRF	vascular endothelial growth factor (VEGF, BMP-2, and ALP). **hPDLCs Proliferation by CCK-8 Assay (On 1st, 3rd and 5th Day), and Migration by Transwell and Scratch Wound Healing Assay (At 24 h):** Proliferation → G3 > G2 > G1 on 3rd and 5th day ($P < 0.05$) Migration → G3 > G2 > G1 ($P < 0.05$) **hPDLCs Mineralization by ALP Assay (On 7th Day) and Alizarin Red Staining (On 14th Day):** G3 > G2 > G1 ($P < 0.05$) **Osteogenic-related Gene Expression by RT-qPCR (on 14th Day):** G3 > G2 > G1 ($P < 0.05$) **Inflammatory- and Osteogenic-related Gene Expression in an Inflammatory Environment by RT-qPCR (on 14th Day):** I-PRF → Significantly down-regulate the expression levels of p65, IL-1β and TNF-α, and significantly up-regulated expression levels of RUNX2, COL1, and OCN when compared to other groups.
Thanasrisuebwong et al. [32] (2020)	Periodontal Regeneration	To evaluate the effect of red/yellow I-PRF on hPDLCs osteogenic differentiation and its behavior relating to the process of mineralized tissue formation.	- hPDLCs	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	Red/Yellow	**Cell Proliferation:** G1) hPDLCs + Growth medium G2) hPDLCs + Red I-PRF G3) hPDLCs + Yellow I-PRF **Cell Migration** G1) hPDLCs + Growth medium G2) hPDLCs + Growth medium + FBS G3) hPDLCs + Red I-PRF G4) hPDLCs + Yellow I-PRF **Osteogenic Differentiation:** G1) hPDLCs + Growth medium G2) hPDLCs + Osteogenic medium G3) hPDLCs + Osteogenic medium + Red I-PRF G4) hPDLCs + Osteogenic medium + Yellow I-PRF	**hPDLCs Proliferation by MTT Assay (At Baseline, and on 3rd and 5th Day):** G2 > G3 > G1 ($P < 0.05$) **hPDLCs Migration by Transwell Assay (At 24 h):** G3 > G4 > G2/G1 ($P < 0.05$) **hPDLCs Mineralization by ALP Assay (On 3rd Day) and Calcification Rate by Alizarin Red Staining (On 7th, 14th, and 21st Day):** Mineralization → G4 > G3 > G2/G1($P < 0.05$) Calcification → G4 > G3 > G2/G1 on 21st day ($P < 0.05$)
Iozon et al.[57] (2020)	Periodontal Regeneration	To evaluate the effect of I-PRF on proliferation and osteogenic differentiation of gMSCs.	- gMSCs	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	Yellow	G1) gMSCs (Control) G2) gMSCs + 5% I-PRF G3) gMSCs + 10% I-PRF G4) gMSCs + 10% I-PRF + FCS	**gMSCs Proliferation by CCK-8 Assay (On 3rd and 7th Day):** Day 3 → G1 = G2 = G4 > G3 ($P < 0.05$) Day 7 → G2 > G1 > G3 > G4 ($P < 0.05$) After three days, gMSCs grown in 10% I-PRF had proliferated significantly less than the other groups. **Osteogenic-related Gene Expression by RT-qPCR (on 14th Day):**

(*continued on next page*)

Table 3 (*continued*)

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
							An overall decrease in the expression of all investigated osteogenic genes was observed in I-PRF-containing media in comparison with the controls. Expression of RUNX2 → G1 = G2 > G3 = G4 (P < 0.05) Expression of SPARC → G1 = G2 = G4 > G3 (P < 0.05)
Fujioka-Kobayashi[45] (2020)	Periodontal Regeneration	To compare the growth factor release of C-PRF with that of I-PRF, and to investigate the regenerative properties of C-PRF and I-PRF on human gingival fibroblasts.	- hGFs	300 g 5 min - (5702 Eppendorf, Hamburg, Germany)	Upper layer	G1) hGFs G2) hGFs + I-PRF G3) hGFs + C-PRF	**Growth Factor Release of PDGF-AA, PDGF-BB, PDGF-AB, TGF-β, VEGF, EGF, and IGF-1 by ELISA (On 1st, 3rd, 7th and 10th Days):** Over 10 days → The increase in the release of PDGF-AA, TGF-β1, and EGF from C-PRF compared with those from I-PRF were the most pronounced. **hGFs Viability by Live/Dead Assay (At 24 h):** Both C-PRF and I-PRF demonstrated excellent cell viability and biocompatibility. **hGFs Migration Assay (At 24 h):** G3 > G2 > G1 **hGFs Proliferation Assay (On 1st, 3rd, and 5th Day):** Day 1 → G3 = G2 = G1 Day 3 → G3 > G2 > G1 Day 5 → G3 > G2 > G1 **Gene Expression by RT-qPCR (On 3rd Day):** TGF-β → G3 = G2 > G1 PDGF → G3 > G1 **Immunohistochemical Staining of COL1 (On 14th Day):** G3 > G2 > G1
Wang et al.[16] (2017)	Periodontal Regeneration	To evaluate the effect of I-PRF on hGFs cultured on smooth and roughened titanium implant surfaces compared to PRP.	- hGFs	700 3 min - (Duo Centrifuge, Nice, France)	Upper layer	G1) hGF + TCP (Control) G2) hGF + TCP + PRP G3) hGF + TCP + I-PRF G4) hGF + PT (Control) G5) hGF + PT + PRP G6) hGF + PT + I-PRF G7) hGF + SLA (Control) G8) hGF + SLA + PRP G9) hGF + SLA + I-PRF	**hGFs Cell Viability by Live/Dead Assay (At 24 h):** Both PRP and I-PRF demonstrated excellent cell viability and biocompatibility. **hGFs Migration Assay (At 24 h):** G3 > G2 > G1 (P < 0.05) G6 > G5 > G4 (P < 0.05) G9 > G8 > G7 (P < 0.05) **hGFs Proliferation by CCK-8 Assay (On 1st, 3rd and 5th Day):** Day 1 → No difference between all groups. Day 3rd and 5th → I-PRF significantly increased cell numbers when compared to the other groups. **hGFs Adhesion Assay by DAPI Staining**

(*continued on next page*)

Table 3 (*continued*)

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
							(At 2, 4, 8 h): No difference between all groups. **hGFs Morphology Assay by Phalloidin-FITC and DAPI Staining (At 8 h):** G3 > G2 > G1 ($P < 0.05$) G6 > G5 > G4 ($P < 0.05$) G9 = G8 = G7 ($P < 0.05$) **Regeneration- and ECM-related Gene Expression by RT-qPCR:** G3 > G2/G1 ($P < 0.05$) G6 > G5/G4 ($P < 0.05$) G9 = G8/G7 ($P < 0.05$) **Immunohistochemical Staining of COL1 (On 7th Day):** G3 > G2 > G1 ($P < 0.05$) G6 > G5 > G4 ($P < 0.05$) G9 > G8 > G7 ($P < 0.05$) Besides, significantly higher total staining was observed on PT surfaces when compared to SLA.
Chai et al.[19] (2019)	Pulp Regeneration	To compare the cellular regenerative activity of hDPCs when cultured with either liquid PRF or traditional PRP.	- hDPCs	700 rpm 3 min - (NR)	Upper layer	G1) hDPCs G2) hDPCs + PRP G3) hDPCs + I-PRF **Inflammatory Environment:** G1) hDPCs G2) hDPCs + LPS G3) hDPCs + I-PRF G4) hDPCs + LPS + I-PRF	**hDPCs Migration by Scratch Wound Healing and Transwell Assay (At Baseline and 24 h), and Proliferation by CCK-8 Assay (On 1st, 3rd and 5th Day):** Migration → G3 > G2 > G1 on 3rd and 5th day ($P < 0.05$) Proliferation → G3 > G2 > G1 ($P < 0.05$) **hDPCs Mineralization by ALP Assay (On 7th Day) and Alizarin Red Staining (On 14th Day):** G3 > G2 > G1 ($P < 0.05$) **Odontogenic-related Gene Expression by RT-qPCR (On 14th Day):** G3 > G2 > G1 ($P < 0.05$) **hDPCs Mineralization by Alizarin Red Staining, and Inflammatory and Odontogenic-related Gene Expression in an Inflammatory Environment by RT-PCR (On 14th Day):** I-PRF → Significantly decreased the expression of inflammatory markers (TNF-a, IL-1b, and p65), and attenuated some of the lost odontogenic potential (expression of Col1a1, DMP-1, and DSPP) induced by LPS. **Immunohistochemical Staining of p65 (On 7th Day):** I-PRF → Significantly reduced the immunofluorescence staining of p65 induced by LPS.
Rafiee et al.[83] (2020)					Upper layer		

(*continued on next page*)

Table 3 (*continued*)

In vitro Studies							
Authors (Year)	Category	Aim of study	Cells/Drugs	I-PRF preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
	Pulp Regeneration and Drug Delivery	To evaluate *in-vitro* drug delivery profile of two differently prepared triple antibiotic-containing I-PRF based scaffolds for pulp regeneration.	**- Triple Antibiotic Mixture:** 1- MET 2- CIP 3- MINO	700 rpm 3 min 4 °C (MF-20R Centrifuge, Awel, China)		G1) I-PRF + Triple Antibiotic Mixture (MET, CIP, MINO) by immersion G2) I-PRF + Triple Antibiotic Mixture (MET, CIP, MINO) by integration G3) I-PRF (Control)	**Chromatographic Method Validation:** Retention Time → MINO (2.3 min), CIP (2.6 min), MET (3.1 min). Maximum UV Absorbances → CIP (268 nm), MET (278 nm), MINO (350 nm) **Drug Release (On 1st, 3rd, 7th, 14th, 21st, and 28th days):** G1 → Burst release within the first 24 h followed by sustained maintenance of all three antibiotics up to 14 days. MINO and MET were still detectable in the 3rd week. G2 → The appropriate characteristics for the sustainable release of the antibiotics weren't observed.
Rafiee et al.[31] (2020)	Pulp Regeneration, Drug Delivery, and Anti-microbial Efficacy	To evaluate the anti-microbial property of an I-PRF scaffold containing triple antibiotic mixture against an Actinomyces naeslundii and Enterococcus faecalis biofilm in an infected immature root canal model.	**- Microbial Biofilm:** 1) Actinomyces naeslundii 2) Enterococcus faecalis **- Triple Antibiotic Mixture**: 1- MET 2- CIP 3- MINO	700 rpm 3 min 4 °C (Eppendorf Centrifuge, Hamburg, Germany)	Upper layer	G1) Biofilm + Triple Antibiotic Mixture (MET, CIP, MINO) G2) Biofilm + I-PRF + Triple Antibiotic Mixture (MET, CIP, MINO) G3) Biofilm + I-PRF G4) Seven-day Biofilm Untreated (Control) G5) Microbial-free Untreated (Control)	**Gene Expression by RT-qPCR:** G2 → The highest antibacterial activity against Actinomyces naeslundii G1 and G2 → Had similar antibacterial property against Enterococcus faecalis G1, G2, and G3 → Revealed higher levels of antibacterial activity against E. faecalis than against Actinomyces naeslundii ($P < 0.001$) **Actinomyces Naeslundii and Enterococcus Faecalis Viability by MTT Assay:** G2 > G1 > G3 ($P < 0.001$)

Abbreviations: I-PRF: Injectable Platelet Rich Fibrin, ATCC: American Type Culture Collection, RPM: Rate Per Minute, Min: Minute, NR: Not Reported, MIC: Minimal Inhibitory Concentration, MBC: Minimal Bactericidal Concentration, PRP: Platelet Rich Plasma, PRF: Platelet Rich Fibrin, ABSM: Allogenic Bone Substitute Material, XBSM: Xenogeneic Bone Substitute Material, RT-qPCR: Reverse Transcription quantitative Polymerase Chain Reaction, A-PRF: Advanced Platelet Rich Fibrin P-PRP: Pure Platelet Rich Plasma, L-PRP: Leukocyte- and Platelet Rich Plasma, IGF-I: Insulin-like Growth Factor-1, VEGF: Vascular Endothelial Growth Factor, PDGF: Platelet-derived Growth Factor, BMP: Bone Morphogenetic Protein, DAPI: 6-diamidino-2-phenylindole, CCK-8: Cell Counting Kit-8, ALP: Alkaline Phosphatase, RUNX-2: Runt-related Transcription Factor 2, OCN: Osteocalcin, COL1: Collagen Type 1, OEC: Outgrowth Endothelial Cell, pOB: Primary Osteoblast, CD: Cluster of Differentiation, ELISA: Enzyme-linked Immunosorbent Assay, ICAM-1: Intercellular Adhesion Molecule 1, hPDLCs: Human Periodontal Ligament Cells, LPS: LPS: Lipopolysaccharide, IL-1b: Interleukin 1 Beta, TNF-a: Tumor Necrosis Factor Alpha, gMSCs: Gingival Mesenchymal Stem Cells, SPARC: Secreted Protein Acidic And Cysteine Rich, hGF: Human Gingival Fibroblast, C-PRF: Concentrated Platelet Rich Fibrin, TCP: Tricalcium Phosphate, PT: Pickled Titanium, SLA: Sand-blasted, Large Grit, Acid-etched, ECM: Extracellular Matrix, hDPCs: Human Dental Pulp Cells, DMP-1: Dentin Matrix Acidic Phosphoprotein 1, DSPP: dentin Sialophosphoprotein, MET: Metronidazole, CIP: Ciprofloxacin, MINO: Minocycline

Table 4
A summary of the included animal studies regarding in the application of I-PRF in regenerative dentistry.

Animal Studies							
Author (Year)	Category	Aim of study	Animals	I-PRF Preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
Elsherbini et al.[14] (2020)	Wound Healing	To evaluate the effects of I-PRF and melatonin on wound healing in diabetic rats.	- 30 diabetic albino rats with surgical defect in their SMGs.	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Upper layer	G1) Control (n = 10) G2) Melatonin (n = 10) G3) I-PRF (n = 10)	**Malondialdehyde Levels Measurement (On 28th Day):** Both I-PRF and melatonin → Reduction of malondialdehyde ($P < 0.05$). **Histological Staining Analysis:** **1) H&E Staining, and 2) Immunohistochemical Staining of Caspase-3, and VEGF (On 28th Day):** - Both I-PRF and melatonin → Reduced caspase-3 and increased vascular endothelial growth factors ($P < 0.05$) → increased SMGs regenerative capacity when compared to diabetic group - Histomorphological structure of SMGs → Melatonin > I-PRF
Mu et al.[29] (2020)	Bone Regeneration, Wound Healing and Anti-inflammatory Efficacy	To assess the angiogenic and osteogenic capacity in rabbit sinus model grafted with DBBM particles soaked in I-PRF.	- 16 New Zealand rabbits (32 Sinuses)	700 rpm 3 min Room temperature (NR)	Yellow	G1) DBBM (n = 16) G2) DBBM + I-PRF (n = 16)	**Micro-CT Findings (At 8th Week Post-implantation):** Bone volume over total volume and thickness of trabecular bone: G1 = G2 ($P > 0.05$) **Laser Confocal Microscopy Photographs of Sequential Fluorochrome Staining (At 2nd, 4th, and 7th Week Post-implantation) together with 2) VG Staining (At 8th Week Post-implantation):** - 2nd week → G2 > G1 ($P < 0.05$) - 4th and 7th weeks → G1 = G2 ($P > 0.05$) - 8th week → Area of new bone formation: G2 > G1 ($P < 0.05$) Area of Bio-Oss: G1 > G2 ($P < 0.05$) **Histological Staining Analysis:** **1) VG Staining and 2) Immunohistochemical Staining of SDF-1 (At 4th Week Post-implantation):** Number of trabecular bones and new bone formation volume, and SDF-1 positive area → G2 > G1 ($P < 0.05$) **3) H&E, TRAP, ALP, and Masson Trichrome Staining (At 4th Weeks Post-implantation):** New bone formation: G2 > G1 ($P < 0.05$)
Yuan et al.[68](2021)	Bone Regeneration	To assess the angiogenesis/ osteogenesis and measure the bone mass reduction using DBBM, GNPs and I-PRF.	- 6 adults male beagle dogs	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	G1) Control G2) DBBM G3) DBBM+I-PRF G4) GNPs G5) GNPs +I-PRF	**Micro-CT Findings (At 8 Weeks Post- implantation):** Highest bone density → G5 **Histological Staining Analysis (At 2nd and 8th Weeks Post-implantation):** **1) H&E, Aniline Blue, and TRAP Staining:** - Angiogenesis and osteogenesis → G1 < G2 < G3 < G4 < G5 ($P < 0.05$) - Osteoclast's activity → G5 had the least activity.
Mu et al.[67] (2020)	Bone Regeneration	To evaluate the effect of I-PRF modified with GNPs for rabbit sinus augmentation.	- New Zealand rabbits	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Yellow	G1) Control G2) GNPs G3) GNPs + I-PRF	**Micro-CT Findings (At 4th and 8th Weeks Post-implantation):** 4th Week → - Bone volume, and trabecular numbers: G3 > G2 > G1 ($P < 0.05$) - Trabecular separation: G1 > G2 > G3 ($P < 0.05$) 8th Week → - Bone volume: G3 > G2 > G1 ($P < 0.05$) - Trabecular number, and trabecular separation:

(*continued on next page*)

Table 4 (*continued*)

Animal Studies							
Author (Year)	Category	Aim of study	Animals	I-PRF Preparation method	Site of I-PRF harvest	Groups	Main Methods and Results
							G3 = G2 = G1 (P > 0.05) **Histological Staining Analysis:** **1) VG Staining (At 4th and 8th Weeks Post-implantation):** 4th Week → New bone area, new bone height, and the angle between the membrane and the implant: G3 > G2 > G1 (P < 0.05) 8th Week → New bone area, new bone height, and the angle between the membrane and the implant: G3 > G2 > G1 (P < 0.05) **2) H&E Staining (At 4th Weeks Post-implantation):** 4th Week → Area of vessels, number of vessels, and vverage area of vessels: G3 > G2 = G1 (P < 0.05)
Aydinyurt et al.[43] (2020)	Periodontal Regeneration	To evaluate the effect of I-PRF in rats with experimental periodontitis.	- 24 Wistar albino rats with ligature-induced periodontitis	3300 rpm 2 min - (NR)	Upper layer	G1) SRP (n = 8) G2) SRP + I-PRF (n = 8) G3) I-PRF (n = 8)	**Histological Staining Analysis:** **1) H&E Staining, and 2) Immunohistochemical Staining of TNF-a, VEGF, IFN-ɤ, and IL-1β (On 31st days):** IL-1β, IFN-ɤ, TNF-α, VEGF, bone resorption, and inflammation values → G1 = G2 = G3 (P > 0.05). **Micro-CT Findings (On 31st days):** - Bone volume, bone levels (mesial/distal) → G1 = G2 = G3 (P > 0.05). - Bone mineral density→ I-PRF > SRP + I-PRF > SRP (P < 0.0001).

Abbreviations: I-PRF: Injectable Platelet Rich Fibrin, SMG: Submandibular Salivary Gland, RPM: Rate Per Minute, Min: Minute, G: Group, H&E: Hematoxylin and Eosin, VEGF: Vascular Endothelial Growth Factor, DBBM: Deproteinized Bovine Bone Mineral, NR: Not Reported, Micro-CT: Micro-Computed Tomography, VG: Van Gieson, SDF-1: Stromal Cell-derived Factor 1, H&E: Hematoxylin and Eosin, TRAP: Tartrate-resistant Acid Phosphatase, ALP: Alkaline Phosphatase, GNP: Gelatin Nanoparticle, SRP: Scaling and Root Planning, TNF-a: Tumor Necrosis Factor Alpha, IFN-ɤ: Interferon Gamma, IL-1b: Interleukin 1 Beta.

Table 5
A summary of the included case reports regarding in the application of I-PRF in regenerative dentistry.

Case Reports							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Intervention	Main Methods and Results
Gasparro et al.[51] (2019)	Wound Healing	To evaluate the effect of I-PRF in the treatment of PCM of the oral cavity refractory to corticosteroid therapy.	- An individual with PCM of the oral cavity refractory to corticosteroid therapy.	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Top layer	I-PRF	**Pain Evaluation by VAS (After 6 Months):** The pain gradually reduced until the score of zero at the fourth infiltration, and the patient remained free of pain during the whole study period. **Clinical Evaluation (After 6 Months):** Clinically, the authors did not obtain a complete healing of the lesion, but a reduced perilesional inflammatory infiltrate was observed at a distance.
Suresh[52] (2021)	Anti-inflammatory Efficacy	To evaluate the successful replantation of an avulsed permanent tooth with an increased extra oral dry time using I- PRF.	- A 21-year-old female with tooth number 11 missing	700 rpm 3 min - (NR)	NR	I-PRF	**Clinical and Radiographic Evaluation (Every 3 Months up to a Year):** The tooth was successfully replanted with no pain and mobility and lesions.
Thanasrisuebwong et al.[50] (2020)	Bone Regeneration	To evaluate the effect of GBR using I-PRF in combination with particulate bone graft and L-PRF for vertical and horizontal bone augmentation prior to implant placement.	- A 55-year-old Asian woman presented with a severe bone defect in posterior mandible	700 rpm 3 min Room temperature (PC-02 Centrifuge, Nice, France)	Red	I-PRF + Bio-Oss + Miner-Oss + L-PRF + Collagen Membrane	**Radiographic Evaluation (After 8 Months):** The result showed a remarkable achievement of vertical and horizontal bone augmentation appropriate for implant placement. **Clinical Evaluation (After 9 Months):** The GBR procedure resulted in good bone quality and quantity of the grafted site. **Histological Staining Analysis (After 9 Months):** **1) H&E Staining:** The histology showed a new normal physiological bone formation in around the implant site as well as at the osteotomy site.
Lorenz et al.[70] (2018)	Bone Regeneration	To evaluate the effect of customized titanium mesh filled with XBSM in combination with A-PRF and I-PRF for reconstruction of a severe tumor-related bony defect in the mandible of a former head and neck cancer patient.	- A 61-year-old female patient affected by squamous cell carcinoma in the anterior floor of the mouth, treated by tumor resection including a block-type resection of the mandible along with bilateral neck dissection	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	Customized titanium mesh + XBSM + A-PRF + I-PRF	**Clinical Evaluation (After 8 Months):** Complete rehabilitation and restoration of the patient's oral function were achieved. **Histological Staining Analysis (After 8 Months):** **1) Azan Staining:** Histological analysis of extracted bone biopsies confirmed that the new bone within the augmented

(*continued on next page*)

Table 5 (*continued*)

Case Reports							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Intervention	Main Methods and Results
							region originated from the residual bone. **Clinical and Radiographic Evaluation (After 4 Months):** The postoperative period was uneventful. The control CBCT scan showed good organization of new bone allowing placement of a dental implant. **Clinical and Radiographic Evaluation (At 15th Month):** significant reduction in pocket depths and significant three-dimensional alveolar bone fill at the treatment site
Chenchev et al.[69] (2017)	Bone Regeneration	To assess the possibility for augmentation of the alveolar ridge in the frontal region of the upper jaw, utilizing a combination of bone graft material, I-PRF and A-PRF.	- A 18-year-old male with expulsion of tooth 11 and partial fracture of the alveolar ridge	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	Bone graft + A-PRF + I-PRF	
Lei et al.[91] (2019)	Periodontal Regeneration	To evaluate the effect of a prefabricated bone construction that was a mixture of blended A-PRF/I-PRF and bone grafts for GBR.	- A 36-year-old male patient diagnosed with severe chronic periodontitis	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	Blended A-PRF + I-PRF + Bio-Oss + Collagen membrane + A-PRF membrane	

Abbreviations: I-PRF: Injectable Platelet Rich Fibrin, PCM: Plasma Cell Mucositis, RPM: Rate Per Minute, Min: Minute, G: Group, VAS: Visual Analog Scale, GBR: Guided Bone Regeneration, H&E: Hematoxylin and Eosin, A-PRF: Advance Platelet Rich Fibrin, NR: Not Reported, XBSM: Xenogeneic Bone Substitute Material, CBCT: Cone Beam Computed Tomography.

[38]'s protocol of 2300 rpm for 15 min, Isik et al. [39]'s method of 2700 rpm for two minutes, and the study by Thanasut et al. [40] which used 1000 rpm for 10 min. Almost all studies on cartilage regeneration, orthodontic tooth movement, pulp regeneration and drug delivery used a protocol of 700 rpm for three minutes for I-PRF preparation. Only Gode et al. [41] centrifuged the tubes at 700 rpm for two minutes and Karci and Baka [42] used a protocol of 800 rpm for three minutes, For periodontal regeneration purposes, all the studies centrifuged blood at 700 rpm for three minutes. Only, Aydinyurt et al. [43] applied a protocol of 3300 rpm for two minutes.

3.2.2. Centrifuge device: Horizontal versus fixed-angle centrifugation

There are two main types of devices used to centrifuge blood for I-PRF preparation: either fixed-angle or horizontal centrifuge devices. While the most common centrifugation system used for PRF preparation yet is the fixed-angle centrifugation system, the horizontal centrifugation system is a more favorable method in research laboratories [44]. The majority of PRP centrifuges historically used horizontal centrifugation, yet few have adapted those protocols for the production of PRF [44]. On this basis, the *in vitro and in vivo* studies on I-PRF included in this paper reported using the fixed-angle method with either Duo, IntraSpin, MF-20R, PC-02, Eppendorf Centrifuge, VE-4000, Ample Scientific Champion F-33D, EBA20, Dynamica Velocity 14R, or TC-SPINPLUS-6 Digital Desktop, except for the study by Fujioka-Kobayashi on I-PRF [45] which used horizontal centrifugation with the Eppendorf Centrifuge.

Although the most common commercially available centrifugation systems are fixed-angled, they are less effective in cell layer separation compared to the horizontal systems [46]. In horizontal systems, the cells are more efficiently separated between the lowest and the highest RCF, leading to the most optimized differentiation of blood contents. Moreover, the cells in fixed-angled centrifugation are more prone to trauma due to the outward-downward force during angled centrifugation systems [44,47]. According to a recent study by Miron et al. [44], the horizontal centrifugation method significantly increases the number of platelets and leukocytes in I-PRF up to 3.5 times more than the fixed-angle centrifugation.

3.2.3. Site of I-PRF harvest

Two types of I-PRF can be attained following centrifugation: red and yellow I-PRF. In the case that the I-PRF is collected merely from the liquid yellow site over the buffy coat, it is referred to as the yellow I-PRF. On the other hand, the sample collected from the red and yellow zone with the buffy coat is considered as the red I-PRF [48]. According to a previous research [48], even minor alterations in the fractionation method can influence the biological and physical properties of the collected sample. Previous studies have reported a higher number of cells (erythrocytes, platelets, and leukocytes) and platelet-derived growth factor (PDGF) for the red I-PRF, and superior fibrin clot formation for yellow I-PRF [48]. Additionally, the viscoelastic properties (clot-forming time, α-angle, and maximal clot firmness) of the yellow I-PRF are substantially greater than that of the red I-PRF due to the above-described variations in cellular components and fibrin network of red and yellow I-PRF [48]. In this regard, Miron et al. [49] have recently introduced a novel methodological approach in order to measure cells and platelets within platelet concentrates in which 100-μL sequential layers were pipetted from approximately 1.2- to 1.5-ml layers above the buffy coat to the red blood cell layer. The results from sequential 100-μL layer in the I-PRF protocol have demonstrated that there was a 3-fold increase in leukocytes and 5- to 6-fold increase in monocytes directly at buffy coat layer in comparison with the baseline. Furthermore, there was a 2.5-fold increase in platelets in all of the aforementioned obtained layers compared to the baseline [49].

Almost all included studies reported investigating the I-PRF obtained from the upper layer of the tube known as the yellow I-PRF.

Table 6
A summary of the included clinical studies regarding in the application of I-PRF in regenerative dentistry.

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
Saglam et al.[54] (2020)	Wound Healing	To compare the effects of I-PRF with those of corticosteroids in the treatment of erosive oral lichen planus.	- 24 Patients with bilateral erosive oral lichen planus	700 rpm 3 min - (IntraSpin, Florida, USA)	NR	G1) Methylprednisolone Acetate (n = 24) G2) I-PRF (n = 24)	**Pain Evaluation by VAS (At Baseline, 1st, 2nd and 6th Months):** G1 = G2 **Quality of Life Evaluation by OHIP-14 (At Baseline, 1st, 2nd and 6th Months):** G1 = G2 **Lesion Size Evaluation (At Baseline, 1st, 2nd and 6th Months):** G1 = G2
Bennardo et al.[53] (2021)	Wound Healing	To compare the efficacy of I-PRF and triamcinolone acetonide injective therapies in patients with symptomatic oral lichen planus.	- 9 patients with symptomatic oral lichen planus	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	Yellow	G1) Triamcinolone Acetonide (n = 9) G2) I-PRF (n = 9)	**Lesion Area Measurement and Morphological Changes Evaluation by Thongprasom Score (At 4th Week):** Lesion extension → G1 = G2 (reduction of 59.8% for G2 and 59.2% for G1) **Pain Evaluation by VAS (At 4th Week):** Pain → G1 = G2 (reduction of 47.6% for G2 and 40% for G1)
Kızıltoprak and Uslu[34] (2020)	Wound Healing	To evaluate the effects of AFG and I-PRF on palatal wound healing and postoperative discomfort.	-36 healthy individuals with the need of FGG	2300 rpm 3 min - (Duo Centrifuge, Nice, France)	Top layer	G1) Control (n = 12) G2) AFG (n = 12) G3) I-PRF (n = 12)	**Wound Healing Evaluation by H_2O_2 Test, MSS, LTH Indices (On 3rd,7th, and 14th days and 1st month), Bleeding Status (On 3rd and 7th days) and Palatal Tissue Thickness (At Baseline, 1st month, and 3rd month):** - Epithelialization on the 14th day→ G1 < G2, G3 ($P < 0.05$) - MSS scores at the 14th day and 1st month→ G2 < G1, G3 ($P < 0.05$) - LTH levels at the 3rd,7th, and 14th day and 1st month → G2 > G1, G3 ($P < 0.05$)

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
							- Bleeding → G1 < G2, G3 (P < 0.05) - Tissue thickness → G1 = G2 = G3 (P > 0.05) **Postoperative Discomfort Evaluation by VAS (on 3rd,7th, and 14th days and 1st month):** VAS at the 7th day → G2 < G1, G3
Nageh et al.[55] (2021)	Anti-inflammatory Efficacy	To evaluate clinically and radiographically the management of internal inflammatory root resorption in permanent anterior teeth using I-PRF.	- 10 healthy patients with 13 anterior teeth diagnosed with internal inflammatory root resorption	700 rpm 3 min - (VE-4000 Centrifuge, Texas, USA)	Upper layer	G1) I-PRF	**Internal Inflammatory Root Resorption and Periapical Lesions Evaluation by CBCT (At Baseline, and 3rd,6th, 12th Months):** The mean volume of internal inflammatory root resorption and periapical lesions decreased (P < 0.05)
Irdem et al.[38](2020)	Bone Regeneration	To investigate the effectiveness of the liquid PRF-DBBM mixture on new bone formation in maxillary sinus augmentation.	- 7 patients with bilateral maxillary sinus atrophy with residual alveolar bone height of 2–5 mm	2300 rpm 15 min - (Ample Scientific Champion F-33D Centrifuge, Georgia, USA)	Yellow	G1) DBBM Alone (n = 7) G2) DBBM + Liquid PRF (n = 7)	**Histological Staining Analysis: Masson's Trichrome Stains + Osteocalcin Antibody (At 4th Week):** No significant difference between the residual crest heights of both groups (P > 0.05) **Radiographic Evaluation by Panoramic Radiography (At 4th Week):** The improvement in both groups looked similar.
Işık et al.[39] (2021)	Bone Regeneration	To evaluate the effect of the screw tent pole technique using particulate allograft with I-PRF on vertical bone augmentation and to compare this with autogenous block bone graft.	- 13 patients with bilateral partial edentulism with insufficient bone height for insertion of short dental implants in	2700 rpm 2 min - (EBA20 Centrifuge, Tuttlingen, Germany)	1 ml upper layer	G1) Autogenous Block Bone Graft (n = 13) G2) Allograft + I-PRF (n = 13)	**Radiographic Evaluation by CBCT (At a Period of 6 Months):** Vertical bone gain → G1 > G2 (P = 0.008) **Histological Staining Analyzing:** **1) H &E Staining**

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
			the posterior of mandible				**(After 6 Months):** Percentage of newly formed bone → G2 > G1 (P < 0.001) **Clinical Evaluation (After 12 months):** The implants' survival rates were 100% in both groups.
Işık et al.[72] (2021)	Bone Regeneration	To assess augmentation success after GBR carried out simultaneously with implant placement using XBSM alone and in combination with liquid PRF.	- 40 partially edentulous patients with residual alveolar bone width of 4–5 mm	700 rpm 3 min - (EBA20 Centrifuge, Tuttlingen, Germnay)	Upper layer	G1) Bovine-derived Xenograft (n = 20) G2) Liquid PRF Enriched Bovine-derived Xenograft (n = 20)	**Radiographic Evaluation by CBCT: 1) Augmentation Thickness (After a Period of 6 Months):** G2 > G1 (P < 0.001) **2) Marginal Bone Loss Level (After 1st and 2nd Years):** G2 < G1 (P < 0.001) **Clinical Evaluation (After 6th month, 1st and 2nd Years):** The implants' survival rates were 100% for both groups.
Thanasut et al.[40] (2021)	Bone Regeneration	To investigate effect of liquid and solid PRF on bone regeneration in repairing alveolar clefts with autologous ABSM.	- 13 patients with 15 alveolar cleft sites	1000 rpm 10 min 25 °C (Dynamica Velocity 14 R Centrifuge, Victoria, Australia)	Yellow	G1) Autologous ABSM (n = 7) G2) Autologous ABSM + Liquid PRF + PRF Membrane (n = 8)	**Radiographic Evaluation by CBCT and Periapical Radiography (At Baseline and After a Period of 6 Months):** No significant difference in regenerated bone volume and density.
Wang et al.[73] (2021)	Bone Regeneration	To test whether or not a digital workflow for GBR with XBSM and I-PRF improved the thickness of the hard tissue compared to the conventional workflow.	- 26 patients with two or three wall horizontal bone defect in the anterior region	700 rpm 3 min - (NR)	Upper yellow layer	G1) XBSM + I-PRF (Conventional) (n = 14) G2) XBSM + I-PRF (Digital) (n = 12)	**Radiographic Evaluation by CBCT (After a Period of 6 Months):-L** abial thickness of hard tissues, and bone gain → G2 > G1 (P < 0.05)- Bone resorption → No significant difference
Wang et al.[74](2021)	Bone Regeneration	To evaluate the impact of different GBR procedures on bone graft contour after wound closure in lateral ridge augmentation.	- 48 patients with 63 augmented sites with a two or three-wall horizontal	700 rpm 3 min - (NR)	NR	G1) XBSM+ Collagen Membrane G2) XBSM+ Collagen Membrane + Healing cap G3) XBSM+ I-PRF + Collagen	**Radiographic Evaluation by CBCT (At a Period of 6 Months):** Labial graft thickness → G3,

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
			bone defect in the anterior region			+ Membrane G4) XBSM+ I-PRF + Surgical Template + Collagen Membrane	G4 > G1, G2 ($P < 0.05$)
Rao et al.[27] (2020)	Bone Regeneration	To evaluate the effect of A-PRF and I-PRF along with iliac bone graft for secondary alveolar bone grafting and compared it with cases in which only iliac bone graft was used.	- 30 patients with alveolar cleft, with age group of ≥ 7 years, having complete unilateral cleft alveolus	700 rpm 3 min - (NR)	Orange superficial layer	G1) Iliac bone graft (n = 15) G2) Iliac bone graft + A-PRF + I-PRF (n = 15)	- For both evaluations the data was clinically favorable in G2. **IOAR Radiographic Evaluation by Bergland Criteria (At 3rd and 6th Month):** G2 > G1 **Periodontal Parameters Evaluation by PPD and Mobility indices (At 3rd and 6th month):** Periodontal status improved in both groups but was more in G2 compared to G1.
Valladão et al.[36] (2020)	Bone Regeneration	To describe the bone gain associated with GBR procedures combining membranes, bone grafts, and PRF (I-PRF and L-PRF) for vertical and horizontal bone augmentation.	- 18 patients who needed vertical or horizontal bone regeneration before installing dental implants	2700 rpm 3 min - (IntraSpin, Florida, USA)	NR	**Horizontal Bone Defect:** G1) ABSM + XBSM + I-PRF + Collagen membrane + L-PRF membrane **Vertical Bone Defect:** G1) ABSM + XBSM + I-PRF + D-PTFE-Ti + L-PRF membrane	**Radiographic Evaluation by CBCT (At Baseline and After a Period of 7.5 ± 1.0 Months):** - The GBR produces an increase in bone thickness ($P < 0.001$) and height ($P < 0.005$). - Bone gain in horizontal defects → Maxilla > Mandible and Anterior > Posterior ($P = 0.014$ and 0.033, respectively) - No difference related to GBR location in vertical defects ($P > 0.05$)
Gülsen and Dereci[71] (2019)	Bone Regeneration	To evaluate the new bone formation after sinus floor augmentation with collagen plugs used as carriers for I-PRF.	- 12 patients who underwent sinus lifting procedures and dental implant placement (18 implants)	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	G1) Collagen Plugs Soaked with I-PRF + Implant	**Radiographic Evaluation by CBCT (After a Period of 6 Months):** - Significant new bone formation at mesial and distal regions of inserted implants ($P < 0.05$) - New bone was

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
							regenerated with I-PRF carried by collagen plugs in sinus floor augmentation.
Kapa et al.[61] (2022)	Periodontal Regeneration	To evaluate the efficacy of sticky bone with I-PRF coated collagen membrane in the treatment of gingival recession.	- 16 patients with isolated Miller's Class I or II recession in the maxillary esthetic zone	700 rpm 3–4 min - (NR)	NR	G1) Sticky Bone (I-PRF + Freeze-dried Bone Allograft) + I-PRF-coated Collagen Membrane	**Radiographic Evaluation by CBCT (At Baseline and After a Period of 6 Months):** Increased labial plate thickness and GT **Periodontal Parameters Evaluation by GT, RD, KTW and CRC (At Baseline and After a Period of 6 Months):** - Increase in GT and KTW and decreased RD and PPD - 12 out of 16 treated cases achieved CRC.
Albonni et al.[62] (2021)	Periodontal Regeneration	To clarify the clinical efficacy of using I-PRF as an adjunctive subgingival irrigation of SRP.	- 15 patients with bilateral periodontal pockets (≥ 5 mm) on a minimum of two teeth on each side	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Yellow layer	G1) Control (n = 338) G2) I-PRF (n = 338)	**Periodontal Parameters Evaluation by PI, PPD, BOP and gingival recession Indices (At Baseline and After a Period of 3rd Months):** No significantly difference in all clinical indices ($P > 0.05$)
Vučković et al.[59] (2020)	Periodontal Regeneration	To evaluate the effect of I-PRF in conjunction with SRP in patients with chronic periodontitis.	- 24 patients with chronic periodontitis who had at least two sites with PPD ≥ 5 mm on contralateral side	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	Upper layer	G1) SRP (n = 24) G2) SRP + I-PRF (n = 24)	**Periodontal Parameters Evaluation by CAL, GML, PPD, BOP, and PI Indices (At 3rd month):** - A significant improvement in investigated clinical parameters for both G1 and G2 ($P < 0.05$) - CAL, GML, PPD, and BOP → G2 > G1 ($P = 0.003$, 0.040, 0.006, and 0.000 respectively). - PI index→ G1 = G2 ($P > 0.05$)
Turer et al.[25] (2020)	Periodontal Regeneration	To determine whether the combined CTG with I-PRF with CAF improved root coverage of deep	- 72 patients with Miller class I and II gingival recession	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	Upper layer	G1) CAF + CTG (n = 36) G2) CAF + CTG + I-PRF (n = 36)	**Periodontal Parameters Evaluation by PPD, CAL, RW, RD, KTH, GT, MRC and CRC**

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
		Miller Class I or II gingival recessions compared with CTG alone with CAF.					**Indices (At Baseline, and 6th month):** - A significant improvement in investigated clinical parameters for both G1 and G2 ($P < 0.05$) - RD and KTH → G2 > G1 ($P = 0.050$ and 0.017, respectively) - PPD, CAL, RW, GT, MRC, and CRC indices → G1 = G2 ($P > 0.05$) **Postoperative Painkiller Assumption, Morbidity and Esthetic Evaluation:** - Significant difference between groups in VAS discomfort ($P = 0.035$) - Patients' and periodontist's VAS evaluation of root coverage, VAS bleeding, and postoperative painkiller consumption → G1 = G2 ($P > 0.05$)
Ozsagir et al.[60] (2020)	Periodontal Regeneration	To evaluate the effect of GT and KTW using I-PRF alone and with MN in individuals with thin periodontal phenotypes.	- 33 systemically healthy patients with thin periodontal phenotypes	700 rpm 3 min - (Duo Centrifuge, Nice, France)	NR	G1) I-PRF (n = 33) G2) I-PRF + MN (n = 33)	**Periodontal Parameters Evaluation by GT and KTW Indices (At Baseline, 1st, 2nd, 3rd, 4th, 5th, and 6th month):** - GT at 6th month→ Significant increase for both G1 and G2 ($P < 0.001$) - KTW at 6th month→G1 = G2 ($P > 0.05$) - However, in the intra-group comparisons, there was a significant difference between GT only in G2 ($P < 0.001$) at the 6th month.
İzol and Üner[17] (2019)	Periodontal Regeneration	To investigate the potential effects of I-	- 40 patients with Miller	700 rpm 3 min	NR		**Periodontal Parameter**

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
		PRF on root coverage of FGG surgery.	class I or II gingival recession	- (NR)		G1) FGG (n = 20) G2) FGG + I-PRF (n = 20)	**Evaluation by Gingival Recession Index (At Baseline and 3rd month):** - At baseline → The mean initial exposed root surface was 4.7 ± 1.49 mm for G2, 4.1 ± 1.07 mm G1, and 4.4 ± 1.31 mm for all subjects. - At 3rd month → The mean root surface coverage values were 3.5 ± 1.05 and 3.9 ± 0.78 mm in the G1 and G2, respectively.
Karadayi and Gursoytrak[76] (2021)	Cartilage Regeneration	To evaluate the efficacy of I-PRF on clinical symptoms of painful TMD, and to determine in which level of diseases, the I-PRF is more effective.	- 36 patients with painful TMDs	700 rpm 3 min - (NR)	Top 3 ml	G1) Arthrocentesis (Wilkes' III) G2) Arthrocentesis (Wilkes' IV) G3) Arthrocentesis (Wilkes' V) G4) Arthrocentesis + I-PRF (Wilkes' III) G5) Arthrocentesis + I-PRF (Wilkes' IV) G6) Arthrocentesis + I-PRF (Wilkes' V)	**TMJ Pain Evaluation by VAS, Dysfunction Evaluation by Helkimo Clinical Dysfunction Index, and MMO Evaluation (At Baseline, Postoperative 10th Day, 30th Day, and 3rd Month):** - VAS and Helkimo Clinical Dysfunction Score → A statistically significant difference in the all controls compared to baseline values - MMO → A statistically significant difference in the 2nd control compared to baseline values
Bera et al.[78] (2021)	Cartilage Regeneration	To evaluate the role of intra articular injection of I-PRF along with arthrocentesis for the management of TMJ-OA.	- 130 patients with OA	700 rpm 3 min - (NR)	NR	G1) Arthrocentesis (n = 67) G2) Arthrocentesis + I-PRF (n = 63)	**TMJ Pain Evaluation by VAS (At Baseline, and After a Period of 3 and 6 Months):** - At Baseline and After 6th Month → G1 = G2 - At 3rd Month → G2 > G1 (P < 0.0001) **MMO Evaluation (After a Period of 6 and 12**

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
							Months): - At 6th Month → G1 = G2 - At 12th Month → G2 > G1 (P < 0.0001)
Ghoneim et al.[77] (2021)	Cartilage Regeneration	To evaluate and compare the efficiency of intra-articular injection of I-PRF following arthrocentesis or arthrocentesis alone in treatment of patients with TMJ disc displacement with reduction.	- 40 patients with reducible anterior disc displacement	700 rpm 3 min - (TC-SPINPLUS-6 Digital Desktop Centrifuge, Oakham, UK)	Top layer	G1) Arthrocentesis (n = 20) G2) Arthrocentesis + I-PRF (n = 20)	**TMJ Pain Evaluation by VAS (After a Period of 6 Months):** G1 > G2 (P < 0.05) **M**MO**, Lateral Movements, and Clicking Evaluation (After a Period of 1 Week, and After 3 and 6 Months):** - MMO, and lateral movements → G2 > G1 (P < 0.05) - Clicking → G1 > G2 (P < 0.05)
Torul et al.[79](2021)	Cartilage Regeneration	To compare the effectiveness of HA and I-PRF in the management of Wilkes stage III internal derangement, and to evaluate the biosupplementation capacity of I-PRF.	- 54 patients with Wilkes stage III internal derangement	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Top layer	G1) Arthrocentesis (n = 18) G2) Arthrocentesis + HA (n = 18) G3) Arthrocentesis + I-PRF (n = 18)	**TMJ Pain Evaluation by VAS and Clicking (At Baseline, After a Period of 1 Week, and After a Period of 1 and 3 Months):** G3 < G1 and G2 (P < 0.05) **M**MO **Evaluation (At Baseline, After a Period of 1 Week, and After a Period of 1 and 3 Months):** G3 > G1 and G2 (P < 0.05)
Yuce and Komerik[75] (2020)	Cartilage Regeneration	To evaluate the effect of intra-articular injection of I-PRF *versus* HA following arthrocentesis in patients suffering from TMJ pain and dysfunction.	- 47 patients (67 TMJs) with internal TMJ derangement	700 rpm 3 min - (NR)	Top layer	G1) Arthrocentesis (n = 16) G2) Arthrocentesis + HA (n = 14) G3) Arthrocentesis + I-PRF (n = 17).[4,5]	-Significant decreases in VAS and increases in MMO values were observed in all 3 groups during 12-month follow-up. **TMJ Pain Evaluation by VAS:** - At 9 months→ G3 < G2 (P < 0.05) **M**MO **Evaluation:** - At 9 and 12 months→ G3 > G2 (P < 0.05)
Gode et al.[41] (2019)	Cartilage Regeneration	To evaluate the effect of I-PRF on the viability of diced	- 40 patients who underwent	2700 rpm 2 min	Upper layer	G1) Diced cartilage (n = 20)	**Cartilage Thickness Measured with**

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
		cartilage, which has been used for dorsum camouflage in rhinoplasty.	open approach primary rhinoplasty	- (NR)		G2) Diced cartilage + I-PRF (n = 20)	**Linear Soft Tissue Ultrasound (At the Postoperative 1st Week and the 3rd Month):** -The mean cartilage graft thickness loss at 3rd month → 0.58 ± 0.21 mm in the G2 and 0.82 ± 0.35 mm in G1 - Volume → Significant loss in G1. - I-PRF was successful in reducing the resorption rate of diced cartilage on nasal dorsum by either increasing the viability or keeping its form.
Albilia et al.[18] (2018)	Cartilage Regeneration	To evaluate the effect of I-PRF in patients with TMJ pain and dysfunction.	- 37 patients (48 TMJs) with painful internal derangement	700 rpm 3 min - (Duo Centrifuge, Nice, France)	Yellow	G1) I-PRF	- 33 of 48 TMJs (69%) showed significant reduction in pain at 8 weeks, and at 3, 6, and 12 months. - The best responders to I-PRF were internal derangement stages Wilkes' IV (78.5%) and V (100%), compared to Wilkes' I (0%), II (47%), and III (33%). **TMJ Pain Evaluation by VAS:** - Significant reductions in pain scores were noted at 8 weeks, and at 3, 6, and 12 months for responders. **Dysfunction and MMO Evaluation:** TMJ Dysfunction and MMO also showed highly favorable trends.
Zeitounlouian et al.[82] (2021)	Orthodontic Tooth Movement	To evaluate the efficacy of I-PRF regarding bone preservation and prevention of root resorption in	- 21 patients with Class II malocclusion with the extraction of the maxillary	700 rpm 3 min - (NR)	NR	G1) Control (n = 21) G2) I-PRF (n = 21)	**Bone Thickness, Bone Height, Root Length, and Dehiscence Evaluation by CBCT (Before**

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
		patients undergoing orthodontic treatment.	first premolars				**and After Retraction):** G1 = G2
Karci and Baka[42](2021)	Orthodontic Tooth Movement	To evaluate and compare the effects of local I-PRF injection and piezocision applications on tooth movement during canine distalization, as well as to evaluate any changes in the periodontal parameters.	- 24 patients with Class II malocclusion with dentoalveolar protrusion or moderate crowding	800 rpm 3 min - (NR)	The middle layer	G1) Control (n = 12) G2) I-PRF (n = 12) G3) Control (n = 12) G4) Piezocision (n = 12)	**Orthodontic Tooth Movements Measurements and Dentoskeletal Changes by Evaluation by CBCT, and Lateral Cephalometric (Before Canine Retraction, and in 12th Week):** G2 = G4 (P > 0.05) **Periodontal Parameters Evaluation by PI, GI & PPD:** G2 = G4 (P > 0.05)
Zeitounlouian et al.[80] (2021)	Orthodontic Tooth Movement	To investigate the effectiveness of I-PRF in accelerating maxillary canine retraction.	- 21 patients with Class II Division 1 malocclusion required the extraction of both maxillary first premolars	700 rpm 3 min - (NR)	Yellow-orange top portion	G1) Control (n = 21) G2) I-PRF (n = 21)	**Orthodontic Tooth Movement Measurement (Before Canine Retraction, at 1st, 2nd, 3rd, 4th, and 5th Month):** - Canine retraction → G2 > G1 at 2nd, 3rd, and 4th month, with the difference being significant at 2nd month (P < 0.05) - Canine rotation and anchorage loss: G1 = G2 (P > 0.05)
Erdur et al.[81] (2021)	Orthodontic Tooth Movement	To evaluate the efficiency of I-PRF in accelerating canine tooth movement and to examine levels of MMP-8, IL-1b, RANKL, and OPG in the GCF during orthodontic treatment.	- 20 patients with Class II Division 1 malocclusion required the extraction of both maxillary first premolars	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	Upper layer	G1) Control (n = 20) G2) I-PRF (n = 20)	**Orthodontic Tooth Movement Measurement (Before Tooth Extraction, in 1st, 4nd, 8rd, and 12th Week):** - Canine tooth movement at all time points→ G2 > G1 (P < 0.001) **GCF Collection (Before Tooth Extraction, in 1st, and 4th Week):** - Stimulation in the levels of inflammatory cytokines→ G2 > G1 (P < 0.001) - The levels of cytokines

(*continued on next page*)

Table 6 (*continued*)

Clinical Studies							
Author (Year)	Category	Aim of study	Participants	I-PRF Preparation Method	Site of I-PRF harvest	Groups	Main Methods and Results
							changed in both groups between 1st and 4th week. - The IL-1b, MMP8, and RANKL values→ G2 > G1 ($P < 0.05$) - The OPG values → G2 < G1 ($P < 0.05$) -The IL-1b, MMP8, and RANKL values→ G2 > G1 ($P < 0.05$) - The OPG values → G2 < G1 ($P < 0.05$)
Karakasali and Erdur[15] (2020)	Orthodontic Tooth Movement	To investigate the efficiency of I-PRF injection on maxillary incisor retraction rate.	- 40 patients with Class II Division 1 malocclusion required the extraction of both maxillary first premolars	700 rpm 3 min Room temperature (Duo Centrifuge, Nice, France)	2–3 ml upper layer	G1) Control (n = 20) G2) I-PRF (n = 20)	**Orthodontic Tooth Movement Measurement (Before Incisor Retraction, in 1st, 2nd, 3rd, and 4th Week):** - The average movements of incisors → G2 > G1 ($P < 0.05$) - No significant difference between the right and left sides in both groups at all time points ($P < 0.05$) -While the movement of incisors was significantly higher in G2 in the week following the PRF injection compared to the other weeks ($P < 0.05$), there were no significant difference in the control group at all-time points ($P < 0.05$).

Abbreviations: I-PRF: Injectable Platelet Rich Fibrin, RPM: Rate Per Minute, Min: Minute, G: Group, VAS: Visual Analog Scale, OHIP-14: 14-item Oral Health Impact Profile, AFG: Autologous Fibrin Glue, FGG: Free Gingival Graft, H2O2: Hydrogen Peroxide, MSS: Manchester Scar Scale, LTH: Landry, Turnbull, and Howley, TMD: Temporomandibular Disorder, TMJ: Temporomandibular Joint, OA: Osteoarthritis, HA: Hyaluronic Acid NR: Not Reported, A-PRF: Advanced Platelet Rich Fibrin, PPD: Periodontal Pocket Depth, GBR: Guided Bone Regeneration, L-PRF: Leukocyte- and Platelet Rich Fibrin, D-PTFE-Ti: Titanium-reinforced Non-resorbable High-density Polytetrafluoroethylene, ABSM: Allogenic Bone Substitute Material, XBSM: Xenogeneic Bone Substitute Material, SRP: Scaling and Root Planning, CAL: Clinical Attachment Loss, GML: Gingival Margin Level, BOP: Bleeding on Probing, PI: Plaque Index, CTG: Connective Tissue Graft, CAF: Coronally Advanced Flap, RW: Recession Width, RD: Recession Depth, KTH: Keratinized Tissue Height, GT: Gingival Thickness, MRC: Mean Root Coverage, CRC: Complete Root Coverage, KTW: Keratinized Tissue Width, MN: Micro Needling, MMO: Maximum Mouth Opening, HA: Hyaluronic Acid, MMP-8: Matrix Metalloproteinase-8, IL-1b: Interleukin 1 Beta, RANKL: Receptor Activator of Nuclear Factor Kappa-light-chain-enhancer of Activated B Cells Ligand, OPG: Osteoprotegerin, GCF: Gingival Crevicular Fluid.

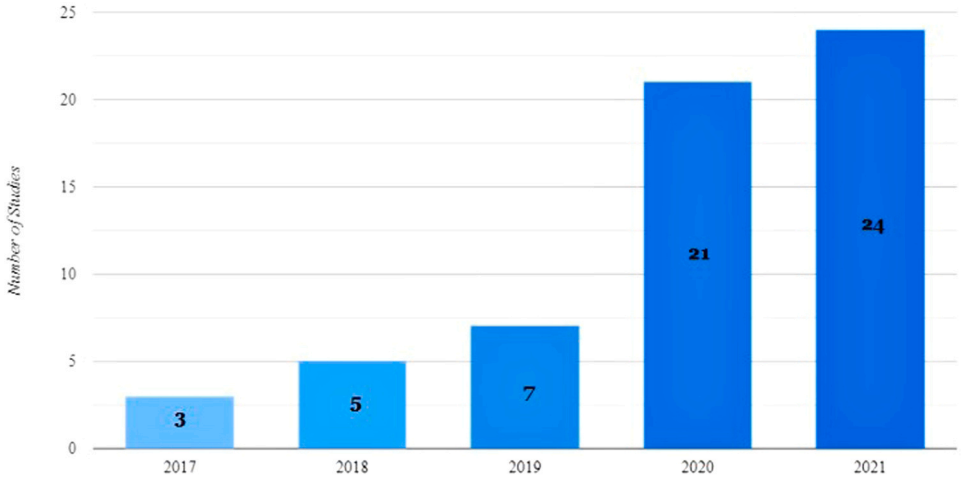

Fig. 2. : Frequency distribution graph. Publication frequency of papers that met the inclusion criteria plotted against year of publication.

Only Thanasrisuebwong et al. [50] reported using red I-PRF in their case study and reported its positive effects on vertical and horizontal bone augmentation. In another study by Thanasrisuebwong et al. [32], the researchers compared the influence of yellow and red I-PRF on human periodontal ligament cells. The authors reported higher cell proliferation and migration for the red I-PRF group compared with the yellow I-PRF group ($P = 0.046$ and $P = 0.024$, respectively). Moreover, significantly increased calcification was reported for the yellow I-PRF compared with the red I-PRF after 21 days ($P = 0.0001$).

3.3. Wound healing and anti-inflammatory efficacy

To date, the investigation of I-PRF on the cells during oral tissue regeneration, inflammation, and wound healing has been evaluated in one *in vitro* [30], two animal studies [14,29], two case reports [51,52], and four clinical studies [34,53–55] (Fig. 3).

3.3.1. In vitro studies on wound healing and anti-inflammatory efficacy

In an *in vitro* study in 2018, Dohle et al. [30] assessed the effect of I-PRF matrices on inducing angiogenesis and wound healing through inflammatory processes in an *in vitro* co-culture. Based on the enzyme-linked immunosorbent assay (ELISA) and Reverse transcription-quantitative polymerase chain reaction (RT-qPCR) tests, an increased expression of factors associated with wound healing (PDGF-BB, intracellular adhesion molecules (ICAM-1), and E-selectin) as well as upregulation of the proangiogenic growth factors (vascular endothelial growth factor (VEGF), bone morphogenetic protein (BMP-2), and alkaline phosphatase (ALP)) for vascular endothelial growth were reported. Thus, the angiogenic activation of human outgrowth endothelial cells was verified following the application of I-PRF.

3.3.2. Animal studies on wound healing and anti-inflammatory efficacy

In 2020, Elsherbini et al. [14] compared the wound healing efficacy of I-PRF with that of melatonin in diabetic rats following a surgical defect in their submandibular salivary glands (SMGs). It was concluded that while both I-PRF and melatonin increased SMGs' regenerative capacity by significantly reducing caspase-3 ($P < 0.001$) and by increasing vascular endothelial growth factors ($P = 0.001$, 0.009 respectively), I-PRF showed inferior results with regards to SMGs' histomorphological structure. In another animal study conducted in 2020, Mu et al. [29] evaluated the angiogenic potential in a rabbit sinus model, which was grafted using deproteinized bovine bone mineral (DBBM) particles dipped in I-PRF. The angiogenic capacity in the I-PRF combined with the DBBM group was reported to be greater than those in the DBBM group. A prolonged-release pattern was reported for the growth factors in the I-PRF combined with the DBBM group for nearly two weeks. The authors [29] reported that the DBBM did not inhibit factor release from I-PRF.

3.3.3. Case reports on wound healing and anti-inflammatory efficacy

In the case study by Gasparro et al. [51] published in 2019, the authors reported that despite a decreased perilesional inflammatory infiltrate after six months of I-PRF injection in a case of plasma cell mucositis, no complete healing of the lesion occurred. It was found that the patient became pain-free after the fourth infiltration, and the visual analog scale (VAS) score remained zero during the entire study. In summary, the authors concluded that while I-PRF showed promising results in accelerating wound healing and decreasing postoperative pain, autologous fibrin presented superior results in wound healing. In a recently published case report, Suresh [52] has evaluated the effect of I-PRF administration in the replantation of an avulsed permanent tooth with an increased extra-oral dry time in a 21-year-old female. The clinical and radiographic evaluations over a year showed that the tooth was successfully replanted with no pain and mobility and lesions.

3.3.4. Clinical studies on wound healing and anti-inflammatory efficacy

Four clinical studies evaluated the effect of I-PRF therapy on wound healing and anti-inflammatory parameters. In the clinical study by Kiziltoprak et al. [34] in 2020, palatal wound epithelialization following I-PRF application was evaluated by H_2O_2 bubbling test and soft tissue healing was assessed by Landry, Turnbull, and Howley (LTH) index. In addition, the Manchester scar scale (MSS), bleeding in palpation, and palatal tissue thickness were investigated for wound healing following the I-PRF application into the palatal area. Higher epithelialization and lower bleeding were found in the I-PRF group compared to the control ($P < 0.05$). However, it was reported that after one month, the autologous fibrin had lower MSS scores and higher LTH levels than the I-PRF groups ($P < 0.05$).

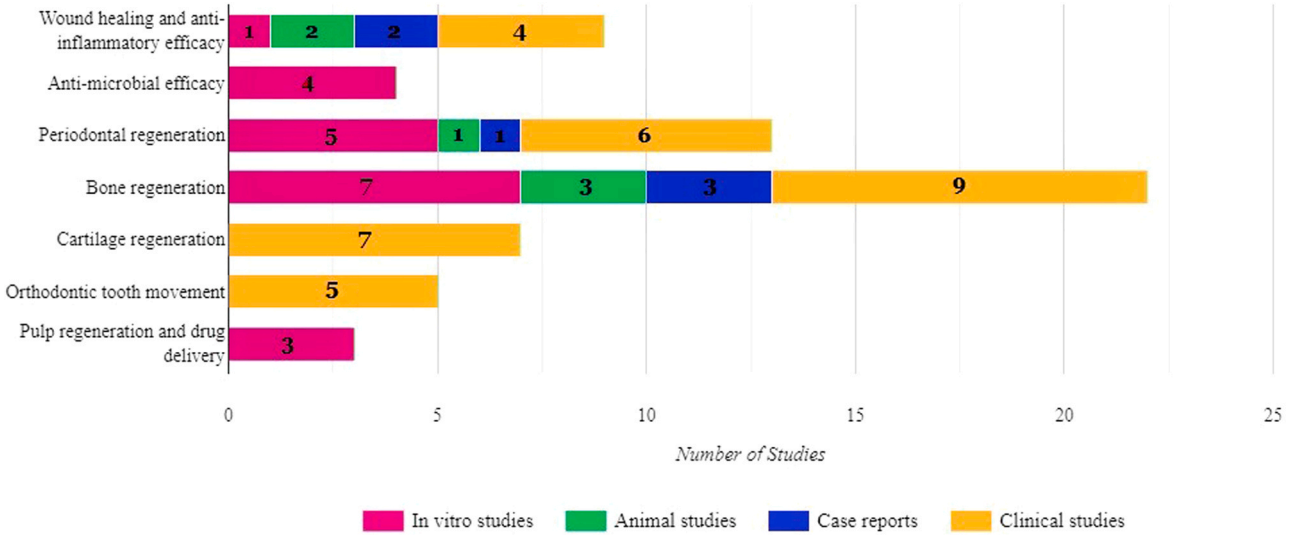

Fig. 3. : Frequency distribution graph. Frequency of the included studies in each category based on their study design.

Significant lower VAS scores were also reported for autologous fibrin compared to I-PRF after a week. No difference was reported between the groups regarding tissue thickness. In 2021, Bennardo et al. [53] carried out research on the efficacy of I-PRF and triamcinolone acetonide injection in patients suffering from symptomatic oral lichen planus. A reduction of 47.6% in the VAS and 59.8% in the extension of lesion was reported for the spots treated with I-PRF, four weeks after the last injections. However, the authors reported no statistically significant differences between I-PRF and triamcinolone acetonide regarding the lesion extension or VAS score. Saglam et al. [54] have recently compared the effects of I-PRF with those of corticosteroids (methylprednisolone acetate) in the treatments of patients with bilateral erosive oral lichen planus over a period of six months. In this regard, one bilateral lesion was injected with I-PRF, whereas the other was injected with methylprednisolone acetate in four sessions at 15-day intervals. Although both groups showed noticeable results in reducing pain and lesion size as well as increasing the quality life of the patients ($P < 0.001$), there were no significant difference in any aforementioned values between these two groups. In another study, Negah et al. [55] have clinically and radiographically investigated the effect of I-PRF administration in the management of root resorption of 10 healthy patients with 13 anterior teeth diagnosed with internal inflammatory root resorption. The clinical evaluation resulted in the resolution of signs and symptoms through the 12-month follow-up period in all of the cases. Moreover, the radiographic evaluation showed a marked decrease in the mean volume of internal inflammatory root resorption and periapical lesions between the preoperative and the 12-month follow-up period.

In general, autologous I-PRF can be suggested as a promising adjunct to the dental and surgical therapeutic processes in increasing the number of growth factors inside the wound and reducing the bacterial count, and helping in wound healing and regeneration.

3.4. Anti-microbial efficacy

The anti-microbial efficacy of I-PRF in the oral environment has been investigated in four *in vitro* studies [24,31,35,56] (Fig. 3).

3.4.1. In vitro studies on anti-microbial efficacy

Jasmine et al. [35] evaluated the I-PRF anti-microbial and anti-biofilm activity against staphylococcus pathogens obtained from a patient with oral abscess. The bactericidal activity of I-PRF against both biofilm-producers and non-biofilm producers was revealed through broth microdilution as minimal bactericidal concentration (MBC) and minimal inhibitory concentration (MIC). A significant decrease in biofilm production was reported at MIC against weak, moderate, and strong biofilm producers ($P < 0.05$). All biofilm-producing bacteria were incapable of biofilm production at MBC concentration. In another work, Rafiee et al. [31] examined the anti-microbial property of I-PRF matrices encompassing a triple antibiotic mixture against *Enterococcus faecalis* and *Actinomyces naeslundii* biofilms in an immature root canal model. The results of gene expression by RT-qPCR revealed that the utmost antibacterial activity against *Actinomyces naeslundii* occurred in the group containing an I-PRF scaffold nurtured with a triple antibiotic mixture. Moreover, the findings of *Actinomyces naeslundii* and *Enterococcus faecalis* viability by MTT assay showed that the maximum and minimum antibacterial efficiency happened in the I-PRF group combined with triple antibiotic and I-PRF alone, respectively. I-PRF scaffold combined with triple antibiotic was shown to substantially reduce live bacteria up to about 92%. In a research by Kour et al. [56], the anti-microbial effect of I-PRF against two periodontal pathogens (*Porphyromonas gingivalis* and *Aggregatibacter actinomycetemcomitans*) was compared with PRP and PRF. The anti-microbial assay by disc diffusion method revealed that, in the case of *Porphyromonas gingivalis*, I-PRF had the widest zone of inhibition, which was significantly wider than that of PRP and PRF ($P < 0.05$). However, in the case of Aggregatibacter actinomycetemcomitans, PRP showed wider zone of inhibition, which was significantly wider than that of PRF and I-PRF ($P < 0.05$). Karde et al. [24] compared the anti-microbial property of I-PRF with other platelet concentrates, such as PRF and PRP obtained from chronic generalized marginal gingivitis patients. It was concluded that I-PRF has maximum anti-microbial efficacy compared with other platelet concentrates, therefore, demonstrating a superior regenerative potential.

In brief, I-PRF application showed favorable anti-microbial efficacy against both periodontal and cariogenic pathogens.

3.5. Periodontal regeneration

A total of five *in vitro* studies [11,32,45,57,58], one animal study [43], one case report [12], and six clinical studies [17,25,59–62] have investigated the influence of I-PRF on periodontal regeneration so far (Fig. 3).

3.5.1. In vitro studies on periodontal regeneration

In 2017, Wang et al. [11] compared the effect of I-PRF and PRP on human gingival fibroblasts cultured on titanium implant surfaces in an *in vitro* setting. According to the authors, both I-PRF and PRP showed excellent gingival fibroblast viability and biocompatibility, and increased migration, spreading, and surface area on tissue culture plastic and pickled titanium, but not acid-etching. I-PRF showed significantly higher transforming growth factor beta (TGF)-β, PDGF, messenger ribonucleic acid (mRNA), fibronectin, and Collagen type 1 compared with PRP. In 2020, Fujioka-Kobayashi et al. [45] compared the regenerative potential of I-PRF and concentrated platelet-rich fibrin (C-PRF) on human gingival fibroblasts. They reported a significant increase in growth factor release for the C-PRF obtained from the buffy coat layer following higher centrifugation protocols compared to the standard I-PRF. Additionally, it was shown that C-PRF had significantly higher gingival fibroblast migration, proliferation, gene expression, and collagen I synthesis. In another *in vitro* study in 2020, Iozon et al. [57] studied the effect of I-PRF on proliferation and osteogenic differentiation of gingival mesenchymal stem cells. The authors reported that while the 5% I-PRF culture significantly increased cell proliferation after seven days, 10% I-PRF significantly reduced cell proliferation. Reduced expression of all osteogenic genes was reported for gingival mesenchymal stem cells in I-PRF cultures. Recently, Zheng et al. [58] and Thanasrisuebwong et al. [32] evaluated the effect of I-PRF on human periodontal ligament cells in their *in vitro* research. Both studies reported enhanced cell proliferation, migration, biological differentiation, and mineralization following conditioning with I-PRF.

3.5.2. Animal studies on periodontal regeneration

In the only animal study carried out on periodontal regeneration capacity of I-PRF, Aydinyurt et al. [43] inspected the efficacy of I-PRF in rats with experimental periodontitis. The researchers claimed positive effects of subgingival I-PRF injection on periodontitis by decreasing bone loss and regulating the inflammatory process. However, according to the authors, the combination of I-PRF injection with scaling and root planning (SRP) did not yield any significant contribution to the treatment of periodontitis.

3.5.3. Case reports and clinical studies on periodontal regeneration

In the case study presented by Lei et al. [12], the effect of advanced platelet-rich fibrin (A-PRF)/I-PRF combined with bone grafts was investigated for guided bone regeneration in a patient with severe chronic periodontitis. Significant alveolar bone gains and reduced pocket depths were reported at the treatment site following a 15-month follow-up. Six clinical studies have so far evaluated the effect of I-PRF therapy on periodontal parameters. In 2019, İzol and Üner [17] examined the effect of I-PRF on root coverage in free gingival graft (FGG) surgery and reported that root surface biomodification with I-PRF promotes root coverage and increases new gingival tissue formation. In another investigation in 2020, Turer et al. [25] researched whether connective tissue graft combined with I-PRF affected root coverage of gingival recessions. The addition of I-PRF to the graft material resulted in a significant reduction of recession depth and an increase in keratinized tissue height compared to the connective tissue graft alone. However, no significant differences were reported between pocket depths, clinical attachment level, recession width, gingival thickness, mean and complete root coverage indices by both periodontal treatments after six months. Vučković et al. [59] compared SRP and I-PRF-added SRP in patients diagnosed with chronic periodontitis. Contrary to the findings of Aydinyurt et al.'s animal study [43], this study revealed that I-PRF combined with SRP significantly improved clinical attachment level, gingival margin level, probing pocket depth, and bleeding on probing indices compared to SRP alone. In another study, Ozsagir et al. [60] compared the effect of I-PRF alone and I-PRF combined with microneedling on gingival thickness and keratinized tissue width in thin periodontal phenotypes. The authors reported that both I-PRF and microneedling-combined I-PRF increased the gingival thickness with the increase being significantly greater in the combined therapy. Kapa et al. [61] have recently evaluated the efficacy of sticky bone with I-PRF coated collagen membrane along with the utilization of coronally advanced flap in the treatment of 16 patients with isolated Miller's Class I or II recession in the maxillary esthetic zone over a period of six months. In this regard, radiographic evaluation by CBCT showed that the labial plate and gingival thickness increased in all cases over that period. Additionally, clinical evaluation demonstrated that all treated cases achieved an increase in gingival thickness and keratinized tissue width as well as a decrease in periodontal pocket and recession depths. Besides, 12 out of 16 treated cases achieved completed root coverage. Albonni et al. [62] have recently assessed the clinical efficacy of administration of I-PRF as an adjunctive subgingival irrigation of SRP in 15 patients suffering from periodontitis with bilateral periodontal pockets (≥ 5 mm) on a minimum of two teeth on each side. Clinical evaluation regarding common periodontal parameters over a period of three months showed no significant differences regarding the adjunctive administration of I-PRF along with the utilization of SRP ($P > 0.05$).

In conclusion, while the *in vitro* studies [11,32,45,57,58] showed promising results in terms of periodontal regeneration potential of I-PRF, there is a lack of consensus on the effect of I-PRF on periodontal parameters evaluated in *in vivo* settings.

3.6. Bone regeneration

A total of 21 studies have so far evaluated the effect of I-PRF on bone regeneration in relation to the oral and maxillofacial structures. These studies are classified under the following subheadings: Seven *in vitro* studies [13,30,37,63–66], three animal studies [29,67,68], three case reports [50,69,70], and nine clinical studies [25,34,36–38,69–72] (Fig. 3).

3.6.1. In vitro studies on bone regeneration

A total of seven *in vitro* studies have evaluated I-PRF effects on bone regeneration using human osteoblasts. Wang et al.*'s in vitro* study [63] on PRP and I-PRF in 2018 was the first research on the effect of I-PRF cultivation on primary human osteoblasts' proliferation, viability, differentiation, mineralization adhesion, and migration. The authors reported that I-PRF resulted in a 3-fold raise in human osteoblast migration in comparison with PRP. In addition, a significantly greater proliferation was induced by I-PRF compared to PRP on the third and fifth day; however, no differences were detected in terms of cell attachment. Furthermore, human osteoblast mineralization by ALP Assay and Alizarin red staining showed significantly higher alizarin red staining at 14 days and ALP staining at

seven days for I-PRF. In an *in vitro* study in 2018, Dohle et al. [30] assessed the effect of I-PRF on human primary osteoblasts in an *in vitro* co-culture, and found lumina and microvessel-like configurations within the I-PRF medium one week following the culture. In 2019, Fernández-Medina et al. [64] compared I-PRF with the other clinical-grade platelet-rich hemoderivatives (A-PRF, pure platelet-rich plasma (P-PRP), leukocyte and platelet-rich plasma (L-PRP)) on osteoblast behavior. The human osteoblast mineralization by Alizarin red staining at day 21 demonstrated superior mineralization properties for I-PRF compared to P-PRP, A-PRF, and L-PRP. However, an I-PRF concentration of > 60% had a detrimental effect on cell viability, metabolic activity, and migration assay. In another *in-vitro* research by Kyyak et al. [13] in 2020, the effect of an allogenic bone substitute material (ABSM) and a xenogeneic bone substitute material (XBSM) with and without I-PRF was assessed on cell characteristics of human osteoblasts. As reported by the authors, the human osteoblast proliferation, attachment, viability, and expression of differentiation and proliferation markers significantly increased in the I-PRF-added bone substitute material (BSM) compared to BSM without I-PRF. Nevertheless, XBSM combined with I-PRF showed inferior results compared to the allogenic bone substitute and ABSM/I-PRF in almost all parameters. To recapitulate, the authors suggested that the use of I-PRF in combination with BSM could enhance the healing process of human osteoblasts. Recently, Kyyak et al. [37] investigated the effect of bovine bone substitute materials (XBSM) combined with I-PRF on the metabolic activity and viability of human osteoblasts. Their findings revealed an increased viability, improved alkaline phosphatase and bone morphogenetic protein 2 expression at earlier phases, and osteonectin expression at later periods when I-PRF was combined with XBSM. In another recent study, Murdiastuti et al. [65] compared the effect of I-PRF and freeze-dried homologous PRP on human osteoblasts and reported that the I-PRF group had the highest number of osteocytes. In 2021, Shah et al. [66] in a study evaluating the effect of osteoblast-like cell line (MG-63) coating of I-PRF on titanium disks, revealed that I-PRF coating of titanium disks resulted in increased proliferation, mineralization, and alkaline phosphatase production.

3.6.2. Animal studies on bone regeneration

Up to now, three animal studies have evaluated the regenerative effects of I-PRF on inducing bone formation. In two studies on I-PRF-induced maxillary bone regeneration, Mu et al. [29,67] assessed the effect of I-PRF modified with gelatin nanoparticles (GNPs) and DBBM for rabbit sinus augmentation. The authors found significantly greater bone creation surrounding the raised Schneiderian membrane for the sinus cavities treated with GNPs-I-PRF hydrogels compared with GNPs gels and the control [67]. Similarly, Mu et al. [29] reported that I-PRF combined with DBBM led to new bone creation in the Schneiderian membrane zone and the basal bone wall. At four weeks, the group treated with GNPs-I-PRF was reported to have significantly higher values for the number of trabecular bones and new bone formation volume [67]. However, lower trabecular separation was reported for GNPs-I-PRF compared with control groups and GNPs. It was concluded that bone resorption was significantly decreased by treating the sinus cavities with GNPs-I-PRF hydrogels [67]. Moreover, Mu et al. [29] concluded that despite the augmented vascular formation and bone remodeling at the early stages of healing using I-PRF incorporated DBBM, the bone volume did not significantly change in a long-term period. Recently, Yuan et al. [68] evaluated the angiogenesis, osteogenesis, and bone mass reduction using deproteinized bovine bone mineral (DBBM), gelatin nanoparticles (GNPs), and I-PRF in male beagle dogs. The researchers showed that the GNPs combined with I-PRF significantly enhanced angiogenesis and woven bone, and reduced osteoclast activity in extraction sockets 2 weeks following the operation. Significant corticalization on the alveolar ridge crest was also reported at 8 weeks post-operation.

3.6.3. Case reports on bone regeneration

To date, three case studies [50,69,70] have evaluated the oral and maxillofacial bone regeneration capacities of bone graft combined with I-PRF. Of these case studies, one assessed bone gain in the maxilla, and the other two examined mandibles. In a case study by Chenchev et al. [69], carried out in 2017, a combination of bone graft material, I-PRF, and A-PRF was used to examine the potential for ridge augmentation in maxillary frontal area. A four-month follow-up with clinical and cone-beam computed tomography (CBCT) scan examinations revealed new bone formation suitable for dental implant placement in the 18-year-old male patient who suffered from tooth 11 expulsion and alveolar ridge partial fracture. In another case reported in 2018, Lorenz et al. [70] investigated the effect of customized titanium mesh filled with XBSM in combination with I-PRF and A-PRF to restore a severe mandibular defect caused by tumor in a 61-year-old head and neck cancer patient. The 8-month clinical and histopathological examination of extracted bone biopsies obtained from the former squamous cell carcinoma patient showed new bone formation in the augmented site. In 2020, Thanasrisuebwong et al. [50] evaluated a combination of I-PRF, particulate bone graft, and leukocyte and platelet-rich fibrin (L-PRF) for horizontal and vertical bone augmentation before implant placement in a patient with severe bone defect in posterior mandible. A 9-month clinical evaluation showed favorable results in bone quality and quantity of the graft site both vertically and horizontally which was appropriate for implant placement.

In summary, all three case reports [50,69,70] assessing the effect of bone graft in combination with A-PRF, I-PRF, and L-PRF substantiated that infiltration with PRF improved the quality of the bone graft material in both maxilla and mandible.

3.6.4. Clinical studies on bone regeneration

Nine clinical studies [27,36,38–40,71–74] have so far investigated oral and maxillofacial bone regeneration following I-PRF application. In a retrospective clinical study conducted in 2019, Gülsen and Dereci [71] examined bone formation following sinus floor elevation with I-PRF carried by collagen plugs. Radiographic evaluation by CBCT showed significant mesial and distal bone formation in the inserted implants after six months ($P < 0.05$). New bone regeneration was detected in sinus floor augmentation using I-PRF, which was carried *via* collagen plugs. In another retrospective clinical study in 2020, Valladão et al. [36] investigated the combination of I-PRF/L-PRF, bone grafts, and membranes for bone augmentation in patients with horizontal or vertical bone defects needing dental implants. Radiographic evaluation by CBCT following 7.5 ± 1.0 months showed that the combination of bone graft with PRF significantly increased bone thickness and height following treatment ($P < 0.001$ and $P < 0.005$, respectively). In a prospective clinical study conducted in 2020, Rao et al. [27] evaluated the effect of A-PRF and I-PRF in combination with iliac bone graft in patients with complete unilateral cleft alveolus. Radiographic evaluation by Bergland criteria and periodontal parameters assessment by periodontal pocket depth (PPD) and mobility indices at third and sixth month revealed more clinically favorable results in the Iliac bone graft + A-PRF + I-PRF

group compared to the patients who only received an iliac bone graft. Generally, all three clinical studies verified the positive effects of injectable PRF application in bone gain as an adjunct to bone graft materials in both maxilla and mandible. In a clinical study by Irdem et al. [38] in 2021, the effectiveness of the DBBM combined with liquid PRF was assessed on new bone formation in patients with bilateral maxillary sinus atrophy in need of maxillary sinus augmentation. It was found that the combination of DBBM with liquid-PRF did not significantly affect new bone formation. Işık et al. [39] compared the effectiveness of particulate allograft combined with I-PRF and autogenous block bone graft on vertical bone augmentation. It was reported that while the particulate allograft material combined with i-PRF is rich in osteoblast cells compared to autogenous block bone graft, it resulted in similar vertical bone gain. In another study by Işık and colleagues [72] on guided bone regeneration simultaneous with implant placement, greater augmentation thickness as well as less marginal bone loss was detected for the bovine-derived xenograft mixed with liquid PRF compared to the xenograft only group.

Thanasut et al. [40] inspected the efficacy of autologous ABSM with and without liquid and solid PRF in bone regeneration in alveolar clefts and found no significant differences in regenerated bone volume and density between autologous ABSM alone and combined with liquid PRF. In a digital workflow for guided bone regeneration using XBSM and I-PRF inspected by Wang et al. [73], a positive effect on the labial thickness of hard tissue was observed with XBSM and i-PRF. Moreover, the authors also investigated the effect of different guided bone regeneration procedures on graft contour in lateral ridge augmentation and found that labial graft thickness was greater when XBSM was combined with I-PRF [74].

In brief, all of the aforementioned *in vitro* and *in vivo* studies showed promising results in terms of bone regeneration following the administration of I-PRF. Only Fernández-Medina et al. [64] reported detrimental effects in cell viability, metabolic activity, and migration assay when the concentration of I-PRF was above 60%.

3.7. Cartilage regeneration

So far, seven clinical studies [18,41,75–79] have investigated the impact of I-PRF application on cartilage regeneration in relation to oral and maxillofacial structures (Fig. 3).

3.7.1. Clinical studies on cartilage regeneration

In 2018, Albilia et al. [18] assessed the effect of I-PRF in patients suffering from temporomandibular joint (TMJ) dysfunction and pain. After eight weeks, and at 3-, 6-, and 12-month follow-ups, the investigators noticed a significant decline in pain scores for responders to intra-articular injections of liquid PRF due to possible remodeling of damaged cartilage surfaces. In a study in 2019, Gode et al. [41] investigated the I-PRF effects on diced cartilage used for rhinoplastic dorsum camouflage. Cartilage thickness measurements at one week and three months postoperative verified the success of I-PRF in decreasing the diced cartilage resorption rate on nasal dorsum by either increasing the viability or maintaining its form. In 2020, Yuce and Komerik [75] compared the effect of I-PRF intra-articular infiltration with that of hyaluronic acid (HA) in patients suffering from TMJ dysfunction and pain. Based on the findings, pain values significantly decreased in the arthrocentesis group combined with I-PRF in comparison to the HA-combined arthrocentesis at nine-month follow-up. Furthermore, maximum mouth opening values in the arthrocentesis group combined with the I-PRF were significantly greater compared to the arthrocentesis group combined with HA at 9 and 12 months postoperatively. In 2021, Karadayi et al. [76] compared the effectiveness of arthrocentesis in combination or without I-PRF for TMJ internal derangement. After three months of follow-up, the authors reported a substantial enhancement in VAS and Helkimo clinical dysfunction scores, as well as the maximum incisal opening of the patients treated with I-PRF-combined arthrocentesis compared to arthrocentesis alone. Recently, Bera et al. [78] assessed the effect of arthrocentesis with intra articular I-PRF injection in the treatment of TMJ osteoarthritis. It was found that while adding I-PRF to arthrocentesis did not alleviate TMJ pain following 6 months of treatment, its repeated injections did positively impact maximal mouth opening. In another study, Ghoneim et al. [77] also compared the efficiency of arthrocentesis with and without intra-articular I-PRF injection in treating TMJ disc displacement with reduction. The authors found significant reduction in click sound and pain intensity and increase in lateral movement and maximal mouth opening when I-PRF was injected. Torul and colleagues [79] examined the effectiveness of I-PRF in the treatment of Wilkes stage III internal derangement and reported that injecting i-PRF following arthrocentesis is more effective than arthrocentesis alone or in combination with hyaluronic acid in the short period.

In conclusion, all seven studies [18,41,75–79] showed favorable trends with regards to cartilage regeneration and temporomandibular disorder treatment following I-PRF therapy.

3.8. Orthodontic tooth movement

To date, five clinical studies [15,42,80–82] have inspected the effect of I-PRF therapy on orthodontic tooth movement (Fig. 3).

3.8.1. Clinical studies on orthodontic tooth movement

In 2020, Karakasali and Erdur [15] assessed the efficiency of I-PRF injection in the retraction rate of the maxillary incisor. the study concluded the movements of incisors were significantly greater in the I-PRF group compared with the control group at all time intervals ($P < 0.05$). In another clinical research by Zeitounlouian et al. [80], the maxillary canine retraction was evaluated following I-PRF therapy. The authors reported a significantly greater canine retraction at the I-PRF side compared with the control site, which was observed only in the second month. Recently, Erdur et al. [81] carried out a research to examine the efficiency of canine tooth movement acceleration following I-PRF treatment and to investigate levels of interleukin 1 beta (IL-1β), matrix metalloproteinase-8 (MMP-8), receptor activator of nuclear factor kappa-B ligand (RANKL), and osteoprotegerin (OPG) in the gingival crevicular fluid during orthodontic treatment. The authors found a significantly increased rate of tooth movement for I-PRF, verified by the stimulation in the inflammatory cytokine levels ($P < 0.001$). In a recent study, Karci et al. [42] compared piezocision with I-PRF injection in tooth movement during canine distalization. It was concluded that while both applications accelerate tooth movement, they do not differ in terms of amount, speed, duration of tooth movement, or periodontal parameters.

In summary, these four clinical studies confirmed the favorable effects of I-PRF on accelerating maxillary anterior tooth movement during orthodontic treatment.

Furthermore, in another recent study by Zeitounlouian et al. [82] on the efficacy of I-PRF in preserving bone and preventing root resorption in orthodontic patients, it was found that I-PRF is not effective in preventing canine root resorption during canine retraction.

In addition, the investigators showed that the prevalence of dehiscence and fenestration was not reduced by I-PRF.

3.9. Pulp regeneration and drug delivery

Up to now, three *in vitro* studies [19,31,83] have evaluated the pulp regeneration and drug delivery potential of I-PRF.

3.9.1. In vitro studies on pulp regeneration and drug delivery

In 2019, Chai et al. [19] compared the cellular regenerative capacity of human dental pulp cells when cultivated with PRP or I-PRF. It was found that I-PRF substantially increases the migration of dental pulp cells compared to PRP. Moreover, a significantly higher alkaline phosphatase activity and expression of genes coding Collagen type 1, dentin matrix acidic phosphoprotein 1 (DMP-1), and dentin sialophosphoprotein (DSPP) were prompted by I-PRF when compared with PRP. In conclusion, it was suggested that I-PRF possess a regenerative potential to stimulate reparative dentin and odontoblastic differentiation in human dental pulp cells. In 2020, Rafiee et al. [83] evaluated the *in vitro* drug delivery profile of two differently prepared Triple Antibiotic-containing I-PRF-based scaffolds for pulp regeneration. The results of I-PRF combined with Triple Antibiotic Mixture (metronidazole (MET), ciprofloxacin (CIP), minocycline (MINO)) verified a burst release within the initial 24 h which sustained for up to 14 days. However, I-PRF combined with Triple Antibiotic Mixture by integration did not show the appropriate characteristics for the sustainable release of the antibiotics. In another drug delivery study by Rafiee et al. [31] in 2020, the efficacy of I-PRF scaffold carrying triple antibiotic mixture was evaluated against *Enterococcus faecalis* and *Actinomyces naeslundii* biofilms in an infected root canal model. It was found that delivering Triple Antibiotic Mixture *via* I-PRF was the most efficient in reducing bacterial metabolic activities when compared with other delivery methods.

In summary, using the 700 rpm for 3 min protocol for I-PRF preparation, all of the studies verified promising results for pulp regeneration and drug delivery potential of I-PRF scaffolds except when combined with Triple Antibiotic Mixture by integration.

4. Future prospects

Although this injectable material is yet to be known by clinicians, there is growing evidence supporting its regenerative potentials. Consequently, to verify the applicability of I-PRF in regenerative procedures, many more intricate and high-quality randomized controlled trials (RCTs) should be performed to demonstrate whether this intervention would improve the clinical outcomes and satisfy the involved clinicians and patients in real clinical situations. Besides, in spite of the limited evidence regarding the effectiveness of I-PRF in other regenerative procedures, the authors believe that the feasibility of I-PRF should be further investigated through rigorous RCTs. In this regard, researchers have recently suggested the possibility of using I-PRF as a promising therapeutic in the treatment of oral mucositis [84].

Very recently, researchers have demonstrated a novel harvesting technique to isolate a concentrated liquid PRF directly from the buffy coat layer superior to the red blood cell layer following L-PRF protocols (2700 rpm for 12 min) [45,49]. They called this obtained liquid PRF as concentrated PRF (C-PRF). In other words, liquid PRF, which is created by a hard spin of blood lacking anticoagulants, forms an upper platelet-poor plasma (PPP) layer, a buffy coat layer (regarded as C-PRF), and the red blood cell layer [87]. In this regard, the findings by Miron et al. have shown that while conventional I-PRF techniques concentrate platelets by 2–3-fold and leukocyte by 1.5-fold from the 1- to 1.2-ml plasma layer compared to baseline concentrations in whole blood, harvesting 0.3–0.5 ml of C-PRF within the buffy coat can increase platelets and leukocytes yield over 10-fold [49].

As discussed earlier, the administration of I-PRF has recently caught extreme attention due to its regenerative, anti-inflammatory, and antibacterial potentials. Furthermore, I-PRF exhibit high cellular contain, growth factors, and cytokines [85]. Since this biomaterial is applied directly to the treatment site, it has high bioavailability. This property makes the growth factors and cytokines affect the treatment site more effectively. In addition, the gradual release of its growth factors and cytokines is another advantage of I-PRF which can significantly regenerate tissues over time [85]. However, I-PRF degrade within a two-week period that limits their long-term application [86]. In order to overcome the aforementioned disadvantage of I-PRF, Fujioka-Kobayashi et al. [86] have lately developed a novel type of PRF called albumin-PRF (Alb-PRF), which is created by combining the liquid PRF layer with the heated albumin layer to form a long-lasting biomaterial capable of releasing growth factors and cytokines for up to 4 months. In fact, it has been shown that heating the platelet-poor plasma forms an albumin gel which generates Alb-PRF when it is mixed back with C-PRF [87]. The lower degradation rate of Alb-PRF in comparison with I-PRF, makes Alb-PRF a more stable biomaterial for longer periods [86]. Since there is limited evidence regarding the prospective relevance of Alb-PRF in regenerative dentistry, the authors deem it necessary that many more preclinical/clinical studies be conducted in order to prove the favorable characteristics of this biomaterial in regenerative procedures. Pre-clinical studies have shown that C-PRF demonstrates an anti-inflammatory activity in murine macrophages and mesenchymal cells, and has a potent inhibitory effect on osteoclastogenesis [87,88]. Although a massive increase in platelets and leukocytes count has been reported for C-PRF compared to the conventional I-PRF [49], no clinical study has investigated the supplementary regenerative value of C-PRF in oral and maxillofacial structures compared to the traditional I-PRF protocol. Thus, further research is suggested to verify the proposed regenerative potential of this novel C-PRF in clinical settings.

5. Conclusion

One of the main goals of platelet concentrate therapy is to a provide a source of growth factors to promote tissue regeneration. Current literature confirms the ability of I-PRF to fulfill this goal and approves the feasibility of I-PRF application as a promising regenerative adjunct to dental procedures. Among the favorable effects of I-PRF are reducing the bacterial count, increasing the amount of growth factors inside the wound, helping with wound healing, and accelerating orthodontic tooth movement as well as periodontal, bone, cartilage, and pulp regeneration. There is a lack of consensus on the effect of I-PRF on periodontal parameters evaluated in *in vivo* settings. Further randomized clinical trials are recommended to validate the proposed benefits.

Funding

None.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknoweldgement

Part of the research reported in this paper was supported by National Institute of Dental & Craniofacial Research of the National Institutes of Health under award number under award number R15DE027533,1 R56 DE029191-01A1, and 3R15DE027533-01A1W1.

The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

References

[1] Saluja H, Dehane V, Mahindra U. Platelet-Rich fibrin: a second generation platelet concentrate and a new friend of oral and maxillofacial surgeons. Ann Maxillofac Surg 2011;1:53–7.
[2] Gaßling VL, Açil Y, Springer IN, Hubert N, Wiltfang J. Platelet-rich plasma and platelet-rich fibrin in human cell culture. Oral Surg, Oral Med, Oral Pathol, Oral Radiol, Endodontol 2009;108:48–55.
[3] Ross R, Glomset J, Kariya B, Harker L. A platelet-dependent serum factor that stimulates the proliferation of arterial smooth muscle cells in vitro. Proc Natl Acad Sci USA 1974;71:1207–10.
[4] Kiran N, Mukunda K, Tilak, Raj T. Platelet concentrates: a promising innovation in dentistry. J Dent Sci Res 2011;2:50–61.
[5] Kawase T, Kamiya M, Kobayashi M, Tanaka T, Okuda K, Wolff LF, et al. The heat-compression technique for the conversion of platelet-rich fibrin preparation to a barrier membrane with a reduced rate of biodegradation. J Biomed Mater Res Part B: Appl Biomater 2015;103:825–31.
[6] Chow TW, McIntire LV, Peterson DM. Importance of plasma fibronectin in determining PFP and PRP clot mechanical properties. Thromb Res 1983;29:243–8.
[7] Marx RE. Platelet-rich plasma: evidence to support its use. J Oral Maxillofac Surg 2004;62:489–96.
[8] Meheux CJ, McCulloch PC, Lintner DM, Varner KE, Harris JD. Efficacy of intra-articular platelet-rich plasma injections in knee osteoarthritis: a systematic review. Arthrosc: J Arthrosc Relat Surg 2016;32:495–505.
[9] Amiri MA, Farshidfar N, Hamedani S. The potential application of platelet-rich fibrin (PRF) in vestibuloplasty. Maxillofac Plast Reconstr Surg 2021;43:1–2.
[10] Choukroun J, Ghanaati S. Reduction of relative centrifugation force within injectable platelet-rich-fibrin (PRF) concentrates advances patients' own inflammatory cells, platelets and growth factors: the first introduction to the low speed centrifugation concept. Eur J Trauma Emerg Surg 2018;44:87–95.
[11] Wang X, Zhang Y, Choukroun J, Ghanaati S, Miron RJ. Behavior of gingival fibroblasts on titanium implant surfaces in combination with either injectable-PRF or PRP. Int J Mol Sci 2017;18:331.
[12] Lei L, Yu Y, Ke T, Sun W, Chen L. The application of three-dimensional printing model and platelet-rich fibrin technology in guided tissue regeneration surgery for severe bone defects. J Oral Implantol 2019;45:35–43.
[13] Kyyak S, Blatt S, Pabst A, Thiem D, Al-Nawas B, Kammerer PW. Combination of an allogenic and a xenogenic bone substitute material with injectable platelet-rich fibrin – a comparative in vitro study. J Biomater Appl 2020;35:83–96.
[14] Elsherbini AM, Ezzat SK. Effect of melatonin versus injectable platelet rich fibrin on critical wound healing in submandibular salivary glands of diabetic rats. J Oral Biol Craniofac Res 2020;10:592–6.
[15] Karakasli K, Erdur EA. The effect of platelet-rich fibrin (PRF) on maxillary incisor retraction rate. Angle Orthodont. 2020;91(2):213–9.
[16] Wang X, Zhang Y, Choukroun J, Ghanaati S, Miron RJ. Behavior of gingival fibroblasts on titanium implant surfaces in combination with either injectable-PRF or PRP. Int J Mol Sci 2017;18:18.
[17] Izol BS, Uner DD. A new approach for root surface biomodification using injectable platelet-rich fibrin (I-PRF). Med Sci Monit: Int Med J Exp Clin Res 2019;25:4744–50.
[18] Albilia J, Herrera-Vizcaino C, Weisleder H, Choukroun J, Ghanaati S. Liquid platelet-rich fibrin injections as a treatment adjunct for painful temporomandibular joints: preliminary results. Cranio: J Craniomandib Pract 2020;38:292–304.
[19] Chai J, Jin R, Yuan G, Kanter V, Miron RJ, Zhang Y. Effect of liquid platelet-rich fibrin and platelet-rich plasma on the regenerative potential of dental pulp cells cultured under inflammatory conditions: a comparative analysis. J Endod 2019;45:1000–8.
[20] Farshidfar N, Amiri MA, Firoozi P, Hamedani S, Ajami S, Tayebi L. The adjunctive effect of autologous platelet concentrates on orthodontic tooth movement: a systematic review and meta-analysis of current randomized controlled trials. Int Orthod 2021;20:100596.
[21] Farshidfar N, Amiri MA, Jafarpour D, Hamedani S, Niknezhad SV, Tayebi L. The feasibility of injectable PRF (I-PRF) for bone tissue engineering and its application in oral and maxillofacial reconstruction: from bench to chairside. Mater Sci Eng: C 2021:112557.
[22] Iozon S, Caracostea GV, Páll E, Şoriţău O, Mănăloiu ID, Bulboacă A-E, et al. Injectable platelet-rich fibrin influences the behavior of gingival mesenchymal stem cells. Rom J Morphol Embryol 2020;61:189–98.
[23] Zhang J, Yin C, Zhao Q, Zhao Z, Wang J, Miron RJ, et al. Anti-inflammation effects of injectable platelet-rich fibrin via macrophages and dendritic cells. J Biomed Mater Res Part A 2020;108:61–8.
[24] Karde PA, Sethi KS, Mahale SA, Khedkar SU, Patil AG, Joshi CP. Comparative evaluation of platelet count and antimicrobial efficacy of injectable platelet-rich fibrin with other platelet concentrates: an in vitro study. J Indian Soc Periodontol 2017;21:97–101.
[25] Ucak Turer O, Ozcan M, Alkaya B, Surmeli S, Seydaoglu G, Haytac MC. Clinical evaluation of injectable platelet-rich fibrin with connective tissue graft for the treatment of deep gingival recession defects: a controlled randomized clinical trial. J Clin Periodontol 2020;47:72–80.
[26] Varela HA, Souza J, Nascimento RM, Araújo RF Jr, Vasconcelos RC, Cavalcante RS, et al. Injectable platelet rich fibrin: cell content, morphological, and protein characterization. Clin Oral Investig 2019;23:1309–18.
[27] Dayashankara Rao JK, Bhatnagar A, Pandey R, Arya V, Arora G, Kumar J, et al. A comparative evaluation of iliac crest bone graft with and without injectable and advanced platelet rich fibrin in secondary alveolar bone grafting for cleft alveolus in unilateral cleft lip and palate patients: a randomized prospective study. J Stomatol, Oral Maxillofac Surg 2020;122(3):241–247–247.
[28] Peters MD, Godfrey CM, Khalil H, McInerney P, Parker D, Soares CB. Guidance for conducting systematic scoping reviews. JBI Evid Implement 2015;13:141–6.
[29] Mu Z, He Q, Xin L, Li Y, Yuan S, Zou H, et al. Effects of injectable platelet rich fibrin on bone remodeling in combination with DBBM in maxillary sinus elevation: a randomized preclinical study. Am J Transl Res 2020;12:7312–25.
[30] Dohle E, El Bagdadi K, Sader R, Choukroun J, James Kirkpatrick C, Ghanaati S. Platelet-rich fibrin-based matrices to improve angiogenesis in an in vitro co-culture model for bone tissue engineering. J Tissue Eng Regen Med 2018;12:598–610.
[31] Rafiee A, Memarpour M, Najibi Y, Khalvati B, Kianpour S, Morowvat MH. Antimicrobial efficacy of a novel antibiotic-eluting injectable platelet-rich fibrin scaffold against a dual-species biofilm in an infected immature root canal model. BioMed Research International 2020;2020:1–8.
[32] Thanasrisuebwong P, Kiattavorncharoen S, Surarit R, Phruksaniyom C, Ruangsawasdi N. Red and yellow injectable platelet-rich fibrin demonstrated differential effects on periodontal ligament stem cell proliferation, migration, and osteogenic differentiation. Int J Mol Sci 2020;21.
[33] Wend S, Kubesch A, Orlowska A, Al-Maawi S, Zender N, Dias A, et al. Reduction of the relative centrifugal force influences cell number and growth factor release within injectable PRF-based matrices. J Mater Sci: Mater Med 2017;28:1–11.
[34] Kiziltoprak M, Uslu MO. Comparison of the effects of injectable platelet-rich fibrin and autologous fibrin glue applications on palatal wound healing: a randomized controlled clinical trial. Clin Oral Investig 2020;24:4549–61.
[35] Jasmine A.T. S, Janarthanan K, Krishnamoorthy R, Alshatwi AA. Antimicrobial and antibiofilm potential of injectable platelet rich fibrin—a second-generation platelet concentrate – against biofilm producing oral staphylococcus isolates. Saudi J Biol Sci 2020;27:41–6.
[36] Amaral Valladao Jr. CA, Freitas Monteiro M, Joly JC. Guided bone regeneration in staged vertical and horizontal bone augmentation using platelet-rich fibrin associated with bone grafts: a retrospective clinical study. Int J Implant Dent 2020;6:72.
[37] Kyyak S, Blatt S, Schiegnitz E, Heimes D, Staedt H, Thiem DGE, et al. Activation of human osteoblasts via different bovine bone substitute materials with and without injectable platelet rich fibrin in vitro. Front Bioeng Biotechnol 2021;9:599224.
[38] Irdem HO, Dolanmaz D, Esen A, Ünlükal N, Şimsek S. Evaluation of the effectiveness of liquid platelet-rich fibrin and deproteinized bovine bone mineral mixture on newly formed bone in maxillary sinus augmentation: a split-mouth. Histomorphometric Study Niger J Clin Pract 2021;24:1366–72.
[39] Isik G, Gunbay T, Uyanikgil Y, Kisaoglu H, Yuce MO. Comparison of autogenous block bone graft and screw tent-pole techniques for vertical bone augmentation in the posterior mandible: a split-mouth randomized controlled study. J Adv Oral Res 2021;12:159–69.
[40] Thanasut A, Silkosessak O, Subbalekha K. Platelet-rich fibrin did not affect autologous bone graft in repairing alveolar clefts. J Oral Maxillofac Surg, Med, Pathol 2021;33:402–7.

[41] Gode S, Ozturk A, Berber V, Kısmalı E. Effect of injectable platelet-rich fibrin on diced cartilage's viability in rhinoplasty. Facial Plast Surg: FPS 2019;35:393–6.
[42] Karcı İÇ, Baka ZM. Assessment of the effects of local platelet-rich fibrin injection and piezocision on orthodontic tooth movement during canine distalization. Am. J. Orthodont. Dentofac. Orthop. 2021;160(1):29–40.
[43] Aydinyurt HS, Sancak T, Taskin C, Basbugan Y, Akinci L. Effects of ınjectable platelet-rich fibrin in experimental periodontitis in rats. Odontology 2020;109(2):422–32.
[44] Miron RJ, Chai J, Zheng S, Feng M, Sculean A, Zhang Y. A novel method for evaluating and quantifying cell types in platelet rich fibrin and an introduction to horizontal centrifugation. J Biomed Mater Res Part A 2019;107:2257–71.
[45] Fujioka-Kobayashi M, Katagiri H, Kono M, Schaller B, Zhang Y, Sculean A, et al. Improved growth factor delivery and cellular activity using concentrated platelet-rich fibrin (C-PRF) when compared with traditional injectable (i-PRF) protocols. Clin Oral Investig 2020;24:24–4383.
[46] Fujioka-Kobayashi M, Kono M, Katagiri H, Schaller B, Zhang Y, Sculean A, et al. Histological comparison of platelet rich fibrin clots prepared by fixed-angle versus horizontal centrifugation. Platelets 2020:1–7.
[47] Lourenço ES, Mourão CFdAB Leite PEC, Granjeiro JM, Calasans-Maia MD, Alves GG. The in vitro release of cytokines and growth factors from fibrin membranes produced through horizontal centrifugation, The in vitro release of cytokines and growth factors from fibrin membranes produced through horizontal centrifugation. J Biomed Mater Res Part A 2018;106:1373–80.
[48] Thanasrisuebwong P, Surarit R, Bencharit S, Ruangsawasdi N. Influence of fractionation methods on physical and biological properties of injectable platelet-rich fibrin: an exploratory study. Int J Mol Sci 2019;20:1657.
[49] Miron RJ, Chai J, Zhang P, Li Y, Wang Y, Mourão CFAB, et al. A novel method for harvesting concentrated platelet-rich fibrin (C-PRF) with a 10-fold increase in platelet and leukocyte yields. Clin Oral Investig 2020;24:2819–28.
[50] Thanasrisuebwong P, Kiattavorncharoen S, Deeb GR, Bencharit S. Implant site preparation application of injectable platelet-rich fibrin for vertical and horizontal bone regeneration: a clinical reportinjectable platelet-rich fibrin forl bone regeneration. J Oral Implantol 2020.
[51] Gasparro R, Adamo D, Masucci M, Sammartino G, Mignogna MD. Use of injectable platelet-rich fibrin in the treatment of plasma cell mucositis of the oral cavity refractory to corticosteroid therapy: a case report. Dermatol Ther 2019;32:e13062.
[52] Suresh N. Entitled "THE MAGIC WAND": a novel treatment option for delayed replantation of an avulsed permanent tooth using injectable platelet-rich fibrin. J Indian Soc Periodontol 2021;25:262–6.
[53] Bennardo F, Liborio F, Barone S, Antonelli A, Buffone C, Fortunato L, et al. Efficacy of platelet-rich fibrin compared with triamcinolone acetonide as injective therapy in the treatment of symptomatic oral lichen planus: a pilot study. Clin Oral Investig 2021;25:3747–55.
[54] Saglam E, Ozsagir ZB, Unver T, Alinca SB, Toprak A, Tunali M. Efficacy of injectable platelet-rich fibrin in the erosive oral lichen planus: a split-mouth, randomized, controlled clinical trial. J Appl Oral Sci: Rev FOB 2021;29:e20210180.
[55] Nageh M, Ibrahim LA, AbuNaeem FM, Salam E. Management of internal inflammatory root resorption using injectable platelet-rich fibrin revascularization technique: a clinical study with cone-beam computed tomography evaluation. Clin Oral Investig 2021;26:1505–16.
[56] Kour P, Pudakalkatti PS, Vas AM, Das S, Padmanabhan S. Comparative evaluation of antimicrobial efficacy of platelet-rich plasma, platelet-rich fibrin, and injectable platelet-rich fibrin on the standard strains of porphyromonas gingivalis and aggregatibacter actinomycetemcomitans. Contemp Clin Dent 2018;9:S325–30.
[57] Iozon S, Caracostea GV, Páll E, Şoriţău O, Mănăloiu ID, Bulboacă AE, et al. Injectable platelet-rich fibrin influences the behavior of gingival mesenchymal stem cells. Rom J Morphol Embryol 2020;61:189–98.
[58] Zheng S, Zhang X, Zhao Q, Chai J, Zhang Y. Liquid platelet-rich fibrin promotes the regenerative potential of human periodontal ligament cells. Oral Dis 2020;26:1755–63.
[59] Vuckovic M, Nikolic N, Milasin J, Djordjevic V, Milinkovic I, Asotic J, et al. The effect of injectable platelet-rich fibrin use in the initial treatment of chronic periodontitis. Srp Arh za Celok Lek 2020;148:280–5.
[60] Ozsagir ZB, Saglam E, Sen Yilmaz B, Choukroun J, Tunali M. Injectable platelet-rich fibrin and microneedling for gingival augmentation in thin periodontal phenotype: a randomized controlled clinical trial. J Clin Periodontol 2020;47:489–99.
[61] Kapa BP, N. KS, G. VG, Mehta DS. Coronally Advanced flap combined with sticky bone and i-PRF-coated collagen membrane to treat single maxillary gingival recessions: Case series. Clinical advances in periodontics. 2021.
[62] Albonni H, El Abdelah Aaad Al, Hamwi OMS, Al Hamoui WB, Sawaf H. Clinical effectiveness of a topical subgingival application of injectable platelet-rich fibrin as adjunctive therapy to scaling and root planing: a double-blind, split-mouth, randomized, prospective, comparative controlled trial. Quintessence Int 2021;52:676–85.
[63] Wang X, Zhang Y, Choukroun J, Ghanaati S, Miron RJ. Effects of an injectable platelet-rich fibrin on osteoblast behavior and bone tissue formation in comparison to platelet-rich plasma. Platelets 2018;29:48–55.
[64] Fernández-Medina T, Vaquette C, Ivanovski S. Systematic comparison of the effect of four clinical-grade platelet rich hemoderivatives on osteoblast behaviour. Int J Mol Sci 2019;20:20.
[65] Murdiastuti K, Olivia N, Kusumadewi WW, Sumito N. In vitro osteogenic potential of freeze-dried homologous platelet-rich plasma. Dent Hypotheses 2021;12:91–5.
[66] Shah R, Thomas R, Gowda TM, Baron TKA, Vemanaradhya GG, Bhagat S. In vitro evaluation of osteoblast response to the effect of injectable platelet-rich fibrin coating on titanium disks. J Contemp Dent Pract 2021;22:107–10.
[67] Mu Z, Chen K, Yuan S, Li Y, Huang Y, Wang C, et al. Gelatin nanoparticle-injectable platelet-rich fibrin double network hydrogels with local adaptability and bioactivity for enhanced osteogenesis. Adv Healthc Mater 2020;9:e1901469.
[68] Yuan S, Li Q, Chen K, Mu Z, Chen T, Wang H, et al. Ridge preservation applying a novel hydrogel for early angiogenesis and osteogenesis evaluation: an experimental study in canine. J Biol Eng 2021;15:19.
[69] Chenchev IL, Ivanova VV, Neychev DZ, Cholakova RB. Application of platelet-rich fibrin and injectable platelet-rich fibrin in combination of bone substitute material for alveolar ridge augmentation – a case report. Folia Med 2017;59:362–6.
[70] Lorenz J, Al-Maawi S, Sader R, Ghanaati S. Individualized titanium mesh combined with platelet-rich fibrin and deproteinized bovine bone: a new approach for challenging augmentation. J Oral Implantol 2018;44:345–51.
[71] Gülşen U, Dereci Ö. Evaluation of new bone formation in sinus floor augmentation with injectable platelet-rich fibrin-soaked collagen plug: a pilot study. Implant Dent 2019;28:220–5.
[72] Işık G, Özden Yüce M, Koçak-Topbaş N, Günbay T. Guided bone regeneration simultaneous with implant placement using bovine-derived xenograft with and without liquid platelet-rich fibrin: a randomized controlled clinical trial. Clin Oral Investig 2021;25:5563–75.
[73] Wang M, Zhang X, Li Y, Mo A. Lateral ridge augmentation with guided bone regeneration using particulate bone substitutes and injectable platelet-rich fibrin in a digital workflow: 6 month results of a prospective cohort study based on cone-beam computed tomography data. Materials 2021;14:6430.
[74] Wang M, Zhang X, Li Y, Mo A. The influence of different guided bone regeneration procedures on the contour of bone graft after wound closure: a retrospective cohort study. Materials 2021;14:583.
[75] Yuce E, Komerik N. Comparison of the efficiacy of intra-articular injection of liquid platelet-rich fibrin and hyaluronic acid after in conjunction with arthrocentesis for the treatment of internal temporomandibular joint derangements. J Craniofacial Surg 2020;31:1870–4.
[76] Karadayi U, Gursoytrak B. Randomised controlled trial of arthrocentesis with or without PRF for internal derangement of the TMJ. J Cranio-Maxillofac Surg 2021;49:362–7.
[77] Ghoneim NI, Mansour NA, Elmaghraby SA, Abdelsameaa SE. Treatment of temporomandibular joint disc displacement using arthrocentesis combined with injectable platelet rich fibrin versus arthrocentesis alone. J Dent Sci 2021;17(1):468–75.
[78] Bera RN, Tiwari P. Evaluating the role of intra articular injection of platelet-rich fibrin in the management of temporomandibular joint osteoarthritis: a STROBE compliant retrospective study. Oral Surg 2021;00:1–6.
[79] Torul D, Cezairli B, Kahveci K. The efficacy of intra-articular injectable platelet-rich fibrin application in the management of Wilkes stage III temporomandibular joint internal derangement. Int J Oral Maxillofac Surg 2021;50:1485–90.
[80] Zeitounlouian TS, Zeno KG, Brad BA, Haddad RA. Effect of injectable platelet-rich fibrin (i-PRF) in accelerating orthodontic tooth movement: a randomized split-mouth-controlled trial. J Orofac Orthop = Fortschr der Kiefer: Organ/ J Dtsch Ges fur Kiefer 2021;82:268–77.
[81] Erdur EA, Karakaslı K, Oncu E, Ozturk B, Hakkı S. Effect of injectable platelet-rich fibrin (i-PRF) on the rate of tooth movement: a randomized clinical trial. Angle Orthod 2021;91:285–92.
[82] Zeitounlouian TS, Zeno KG, Brad BA, Haddad RA. Three-dimensional evaluation of the effects of injectable platelet rich fibrin (i-PRF) on alveolar bone and root length during orthodontic treatment: a randomized split mouth trial. BMC Oral Health 2021;21:92.
[83] Rafiee A, Memarpour M, Taghvamanesh S, Karami F, Karami S, Morowvat MH. Drug delivery assessment of a novel triple antibiotic-eluting injectable platelet-rich fibrin scaffold: an in-vitro study. Curr Pharm Biotechnol 2020;22(3):380–8.
[84] Amiri MA, Farshidfar N, Hamedani S. The prospective relevance of autologous platelet concentrates for the treatment of oral mucositis. Oral Oncol 2021;122:105549. (-).
[85] Miron RJ, Fujioka-Kobayashi M, Hernandez M, Kandalam U, Zhang Y, Ghanaati S, et al. Injectable platelet rich fibrin (i-PRF): opportunities in regenerative dentistry? Clin Oral Investig 2017;21:2619–27.

[86] Fujioka-Kobayashi M, Schaller B, Mourão CFDAB, Zhang Y, Sculean A, Miron RJ. Biological characterization of an injectable platelet-rich fibrin mixture consisting of autologous albumin gel and liquid platelet-rich fibrin (Alb-PRF). Platelets 2021;32:74–81.

[87] Kargarpour Z, Nasirzade J, Panahipour L, Miron RJ, Gruber R. Liquid PRF reduces the inflammatory response and osteoclastogenesis in murine macrophages. Front Immunol 2021;12:636427.

[88] Kargarpour Z, Nasirzade J, Panahipour L, Miron RJ, Gruber R. Platelet-rich fibrin decreases the inflammatory response of mesenchymal cells. Int J Mol Sci 2021;22:22.

[91] Lei L, Yu Y, Ke T, Sun W, Chen L. The application of three-dimensional printing model and platelet-rich fibrin technology in guided tissue regeneration surgery for severe bone defects. J Oral Implantol 2019;45:35–43.