Journal of Population Therapeutics & Clinical Pharmacology

RESEARCH ARTICLE
DOI: 10.47750/jptcp.2023.30.17.023

PRF: EMPOWERING PERIODONTAL REGENERATION FROM WITHIN

Dr. Ruparani Bodduru[1]*, Dr. Aiysha Nudrath[2], Dr. Sushmitha Hari[3], Dr. Venkata Aruna Mushti[4], Dr. Deepika Veldurthi[5], Dr. Sindhe sai charan[6]

[1*]Professor & HOD, Department of Periodontics and Implantology, MNR Dental College and Hospital, Sangareddy, Telangana, India.
[2,3,4]III Year PG student, Department of Periodontics and Implantology, MNR Dental College and Hospital, Sangareddy, Telangana, India.
[5]Reader, Department of Periodontics and Implantology, MNR Dental College and Hospital, Sangareddy, Telangana, India.
[6]Senior Lecturer, Department of Periodontics and Implantology, MNR Dental College and Hospital, Sangareddy, Telangana, India.

[1]*Email: drrupaprakash@gmail.com,
[2]Email: nudrathaiysha96@gmail.com,
[3]Email: sushmi.hari94@gmail.com,
[4]Email: arukiranjonnalagadda@gmail.com,
[5]Email: deepikaa@gmail.com,
[6]Email: dr.saicharanshindhe28@gmail.com

***Corresponding Author: Dr. Ruparani Bodduru**
*Professor & HOD, Department of Periodontics and Implantology, MNR Dental College and Hospital, Sangareddy, telangana, India. Email id: drrupaprakash@gmail.com

Abstract
In periodontology, platelet-rich fibrin (PRF) has become a useful add-on therapy with the goal of accelerating periodontal tissue regeneration and repair. PRF is a concentrated form of platelets, growth factors, and cytokines, an autologous preparation made from the patient's own blood. Due to its distinctive makeup, PRF can successfully encourage tissue regeneration and healing after a variety of periodontal procedures. It can enhance the results of treatments like guided tissue regeneration and bone grafting by acting as a natural scaffold to assist with the growth of new bone and connective tissue. PRF has been shown to accelerate soft tissue healing, lessen postoperative pain, and reduce swelling and inflammation. Its use in procedures including implant placement, gingival recession coverage, and flap surgeries have demonstrated enhanced wound healing and aesthetic results. The ability of PRF to create an advantageous milieu rich in cytokines and growth factors aids in improving the results of different periodontal procedures.

Keywords: - Platelet-rich fibrin (PRF), Periodontics, Periodontal regeneration

INTRODUCTION

Periodontal disease is a complex, multifaceted illness characterized by connective tissue attachment loss and periodontal damage. Periodontal therapy targets to reduce inflammation, prevent the progression of periodontal disease, and repair damaged periodontal tissues. Periodontal regeneration

is a complex multifactorial process that orchestrates physiologic events such as cell migration, adhesion, differentiation, and proliferation.[1] Bone grafts, soft tissue grafts, guided tissue regeneration, root biomodifications, and combinations of these procedures are examples of periodontal regenerative techniques.[2] According to current thinking, regenerative periodontal therapies can only restore a percentage of the original tissue volume[2] and have a limited possibility for total periodontal restoration.[3] In addition, many biomaterials have been employed for periodontal tissue regeneration.
Interactions between epithelial cells, gingival fibroblasts, periodontal ligament cells, and osteoblasts are required for periodontal wound healing. During wound healing, vascular disruption causes fibrin formation, platelet accumulation, and the release of multiple growth factors into tissues from platelets[4] via molecular signals mediated mostly by cytokines and growth factors. There is evidence that platelets embrace growth factors and cytokines that play a significant role in inflammation and wound repair.[5] Platelets also release fibrin, fibronectin, and vitronectin, which serve as connective tissue matrix and adhesion molecules for more efficient cell movement.6This has given rise to the concept of employing platelets as therapeutic agents to enhance tissue repair, notably in periodontal wound healing.
Platelets' regenerative potential was introduced in the 70s[7]; it was observed that they comprise growth factors that are liable for boosted collagen production, blood vessel growth, cell mitosis, recruitment of other cells that relocate to the site of injury, and cell differentiation induction, among others.[8]
The use of platelet concentrates for in vivo tissue engineering applications is one of the most recent innovations in oral surgery:

1) Platelet-rich plasma (PRP) and
2) Platelet-rich fibrin (PRF). Platelet concentrates are a concentrated dispersion of platelet growth factors that are bioactive surgical additives used locally to promote wound healing.[9]

Whitman et al.[9] were the first to employ platelet-rich plasma in oral surgical operations, demonstrating significant benefits because it stimulates osteoprogenitor cells in the host bone and bone transplant. However, there is a risk of utilizing it because bovine thrombin, which is used to handle PRP, can create antibodies against factors V, XI, and thrombin, which can cause coagulopathies that can be fatal.[8]
PRF, on the other hand, was initially used by Choukroun et al.[10] in 2001 and is now considered a new generation of platelet concentrate, made up of an autologous fibrin matrix[11] and having various advantages over PRP, including quicker preparation and the absence of chemical modification.
Platelets constitute the second most abundant corpuscles in the blood[12], consisting of cytoplasmic fragments that lack a core derived from megakaryocytes.[13,14] With a lifespan of 7 to 10 days[14] and a normal peripheral blood concentration of 150-450 109/L[12] are often biconvex discoid structures shaped like lenses with dimensions of roughly 2.0-4.0 by 0.5m and a mean volume of 7-11 fl.[12.]
Platelets formed in the periphery correspond to a phospholipid membrane, a series of microtubules. A very extensive canalicular system connects the surface to the cytoplasm[15] with the most important elements (leukocytes and growth factors, the latter of which are polypeptides) to achieve healing and repair processes that participate in differentiation migration, proliferation, and cell metabolism.[16.]
Platelet-derived growth factors activate and recruit stem cells to the site of damage, boosting cell mitosis and triggering angiogenesis and osteogenesis[17] have also revealed periosteum cell mitogenic response to bone repair.[18] Platelets also generate cytokines and are responsible for shaping platelet activation and leukocyte proliferation and differentiation, playing a vital role in immunology, specifically in the inflammatory mechanism.[17]
Furthermore, fibrin, as a bridge molecule, allows for a sequence of cell contacts and acts as a provisional matrix in which cells can proliferate, organize, and carry out their duties, primarily in areas of injury or inflammation[19], creating a substrate for fibroblast and endothelial cell movement, important in angiogenesis and tissue repair.

PLATELET-RICH FIBRIN

PRF is an autologous leukocyte-platelet-rich fibrin matrix[11,17], possessing a tetra molecular structure that includes platelets, cytokines, and stemcells[20,21] and serves as a biodegradable scaffold[22] that fosters micro vascularization and can direct epithelial cell migration to its surface.[21,23] Additionally, PRF may act as a vehicle for transporting cells involved in tissue regeneration[24] and tends to have an extended release of growth factors [25] over 1 to 4 weeks, boosting the environment for wound healing in a significant time.[26] It is slowly remodeled and has a complex architecture of a strong fibrin matrix with desirable mechanical properties analogous to a blood clot.[26]

Some studies[27-30] have proven that PRF is a healing biomaterial with tremendous potential for bone and soft tissue regeneration, lacking inflammatory reactions, and may be used alone or in conjunction with bone grafts, promoting hemostasis, bone growth, and maturation. In vitro, investigations revealed that this autologous matrix has a high potential to enhance cell attachment[24] and osteoblast proliferation and differentiation.[29] According to Dohan et al.[21,30], PRF possesses immunological and antimicrobial capabilities, can cause leukocyte degranulation, and contains certain cytokines that stimulate angiogenesis and pro/anti-inflammatory responses.

The difference between a natural blood clot and PRF is that the latter is more homogeneous, stable, and easy to handle and place in the indicated local area.[31]

CLASSIFICATION OF PLATELET-RICH CONCENTRATES

Dohan Ehrenfest et al.[32,33] in 2009, proposed the first classification, which is now widely accepted. The classification is simple, based on the presence or absence of leukocytes and the fibrin architecture density in platelet concentrates. Based on the differences in these criteria, it can be categorized into four major types: pure platelet-rich plasma, pure platelet-rich fibrin (PRF), leukocyte and platelet-rich plasma, and leukocyte and PRF, which are detailed in figure 1.

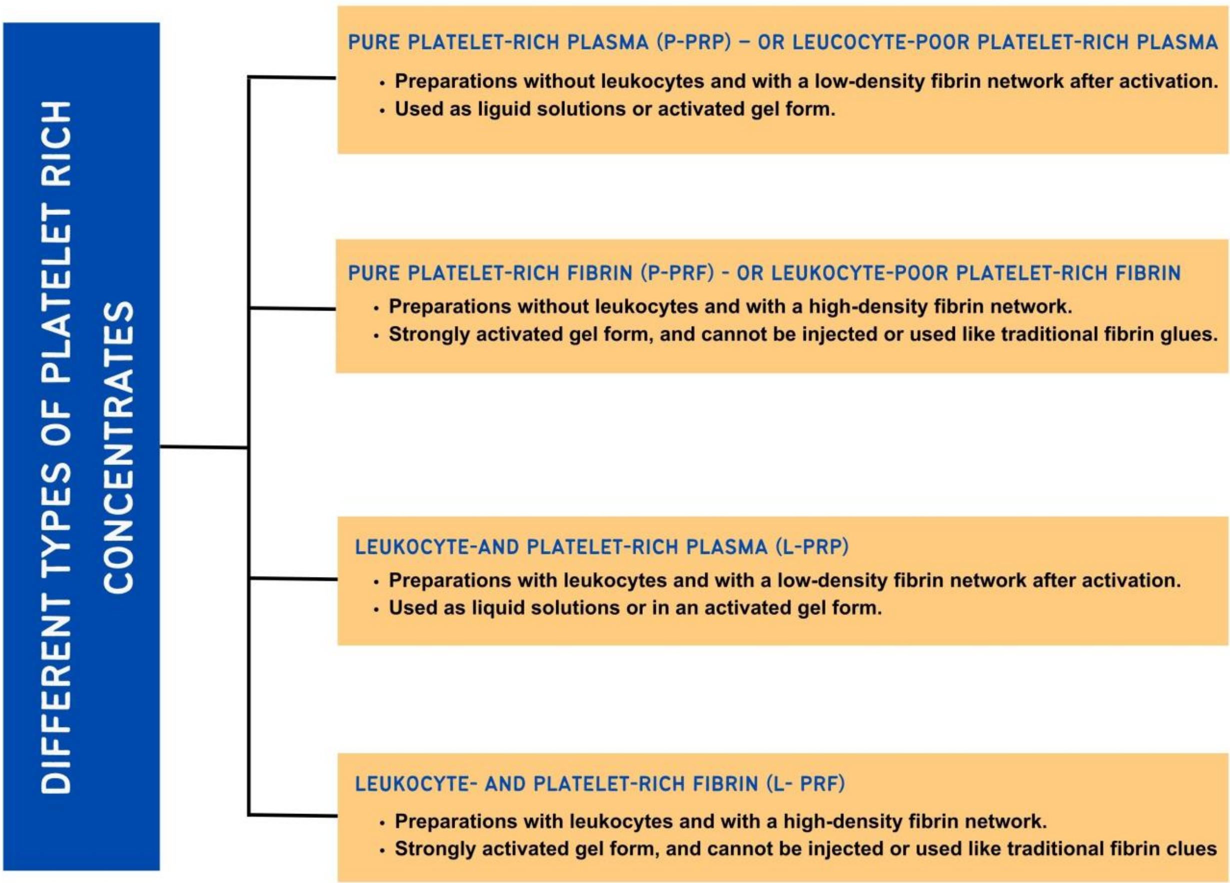

Figure 1- Description of different types of platelet-rich concentrates

Pure Platelet-rich Fibrin (P-PRF) is a new generation of platelet concentrates that uses a simplified approach and does not require biochemical blood handling. Dr. Joseph Choukroun directed the PRF research. [34, 35]

To obtain the correct amount and quality of fibrin matrix, platelets, and growth factors, a consistent technique for PRF production should be followed.

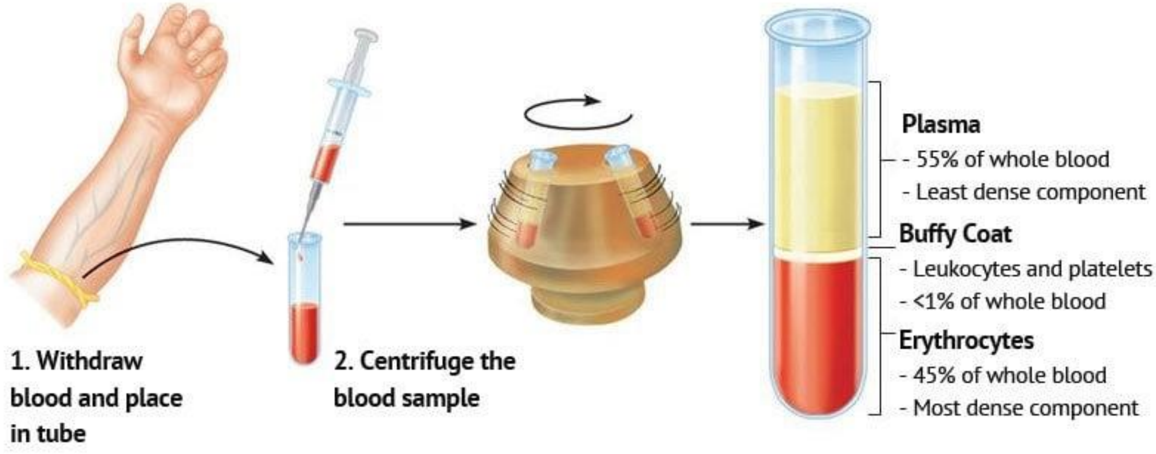

Figure 2 - Platelet Concentrate

A needle is used for 10mL blood collection in sterile glass-coated plastic tubes without anticoagulants that are directly centrifuged at 3000 revolutions per minute for ten minutes once the blood comes into contact with the test tube.

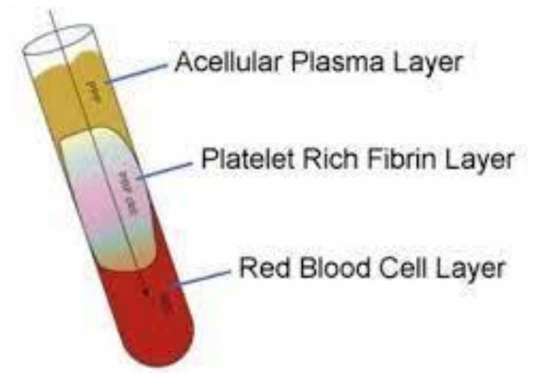

Figure :3 Layers obtained after centrifugation

(Courtesy: Mohan SP, Jaishangar N, Devy S, Narayanan A, Cherian D, Madhavan SS. Platelet-rich plasma and platelet-rich fibrin in periodontal regeneration: a review. Journal of pharmacy & bioallied sciences.)

The uppermost layers of the test tube are made up of one-celled PPP (platelet deficient plasma), RBCs at the bottom, and a PRF clot.

The fibrin clot formed when action was eliminated from the tube, and therefore the tangled-up red blood cells scraped off and discarded.

PRF can even be formed as a **membrane** by compressing and draining the fluids trapped within the fibrin clot.[36,37] wall, the platelet is activated, and the action cascade is initiated. Types of PRF and their mode of preparation [38] are given in Table 1

TABLE -1 – Types of PRF

	Centrifugal speed	Centrifugal time	Tube type	Nature of the obtained PRF
Platelet rich fibrin (L-PRF)	2700rpm	12 mins	glass tube	solid
Titanium platelet-rich fibrin (T-PRF)	2700rpm	12mins	titanium tube	solid
Advanced platelet-rich fibrin (A-PRF)	1300rpm	14mins	glass tube	solid
Albumin platelet-rich fibrin (Alb-PRF)	1300rpm	8 mins	glass tube	solid
Injectable platelet-rich fibrin (I-PRF)	700rpm	3 mins	plastic tube	liquid

ADVANTAGES OF PRF [39]

- Completely an autologous product
- Limits blood manipulation while avoiding biochemical manipulation
- As polymerization occurs naturally, no bovine thrombin is required.
- PRF fibrin matrix contains growth factors, leukocytes, and cytokines that aid in healing.
- When compared to other platelet concentrates, it exhibits prolonged growth factor release.
- The PRF membrane is extremely flexible and elastic.
- Low-cost, easy method that requires only one centrifugation stage.

SHORTCOMINGS OF PRF [39]

- The success of PRF preparation is primarily determined by the speed with which blood is handled
- Considering the structural integrity of PRF membrane changes over time, it should be employed immediately.
- Due to the risk of bacterial infection and dehydration, storing PRF membrane is impossible.
- Being an Autologous product, the quantity of PRF is limited and cannot be used in general surgery.

PROPOSED MECHANISM OF ACTION

Choukroun's PRF is based on mechanical concentration. Hence, the properties of platelet concentrates depend on the procedures used.[40,41] PRF is a platelet-derived condensation of suspended growth factors[42-45] These growth factors are tissue regeneration enhancers that have ramifications in wound healing. PRF clinical utilization can be optimized using elaborated growth factors from PRF.[46-47]

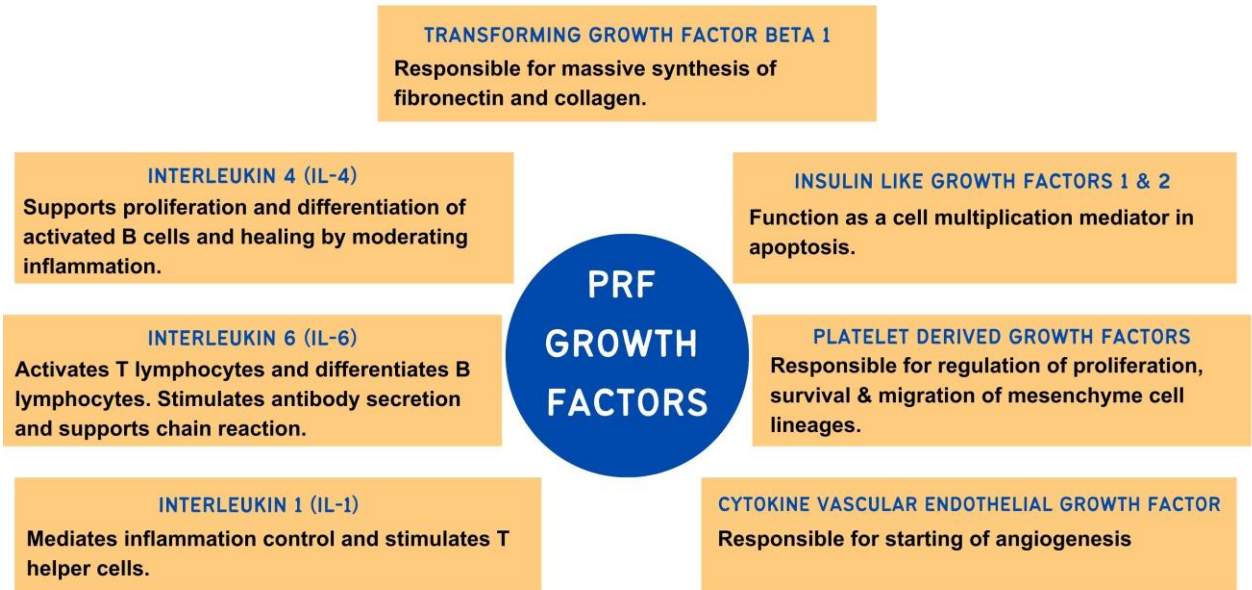

Figure – 4 Role of platelet-rich fibrin growth factors and cytokines in tissue regeneration and wound healing[48]. Transforming growth factor ß1, insulin-like growth factors 1 and 2, platelet-derived growth factor, cytokine vascular endothelial growth factor, and interleukin 1, 4, and 6

(Courtesy - Verma UP, Yadav RK, Dixit M, Gupta A. Platelet-rich fibrin: a paradigm in periodontal therapy–a systematic review. Journal of international society of preventive & community dentistry. 2017 Sep;7(5):227.)

PRF IN PERIODONTICS

In surgical procedures, PRF could be used as a resorbable membrane for guided bone regeneration (GBR) [49], preventing undesirable cells from migrating into the bone defect and providing a space for osteogenic and angiogenic cells to migrate and the underlying blood clot to mineralize.[50]
Simonpieri et al.[51] introduced the notion of "natural bone regeneration" (NBR), which comprises bone volume and gingival tissue regeneration via PRF membranes.
PRF membrane has shown promising clinical outcomes in the treatment of periodontal infrabony defects[49], in the protection of open wounds from the oral environment when sutures cannot bind the mucosal borders[17,21,49,51], and in the acceleration of hard and soft tissue healing.[21,51,52] PRF membrane was utilized as the only grafting material in a few clinical investigations53,54 to achieve maxillary sinus floor augmentation, with encouraging outcomes.
Tofler et al.[55] suggested using a PRF to seal a previously undiscovered sinus membrane perforation during a lateral window osteotomy in a maxillary sinus lift technique.
PRF membrane promotes soft tissue repair and wound healing by protecting the surgical site.[52,55] When mixed with bone graft, it may act as a "biological connector," attracting stem cells, promoting the migration of osteoprogenitor cells to the center of the graft, and providing neo-angiogenesis.[55]
Furthermore, the inclusion of PRF in the bone transplant can reduce the volume of bone replacement utilized and appears to promote graft revascularization by boosting angiogenesis.[56]
Simonpieri et al.[53] proposed employing a PRF-bone graft mixture to fill bone deficiencies or, in the event of immediate implants, covering it with many PRF layers, with good clinical results.
Yilmaz et al.[57] evaluated the histological and stereological healing effects of -TCP and PRF alone and in combination in standardized bone defects in pig tibiae. When -TCP and PRF were administered together, the newly produced bone was much greater than when utilized separately.
Furthermore, PRF may act as a biological adhesive to hold the particles together, making manipulation of the bone grafts easier.[57]

CONCLUSION

In vitro and in vivo investigations on using PRF alone or combined with other biomaterials have yielded safe and encouraging outcomes, with no contradicting findings. It offers various advantages and potential applications in medicine and dentistry. Platelet-rich fibrin is a well-accepted, minimally invasive method with low risks and good clinical outcomes.

REFERENCES

1. W.V. Giannobile-The potential role of growth and differentiation factors in periodontal regeneration. J Periodontol, 67 (1996), p. 545e53
2. H. Greenwell-Committee on research, science and therapy, American Academy of Periodontology. Position paper: guidelines for periodontal therapy. J Periodontol, 72 (2001), pp. 1624-1628
3. L. Sander, T. Karring. Healing of periodontal lesions in monkeys following the guided tissue regeneration procedure. A histological study. J Clin Periodontol, 22 (1995), pp. 332-337
4. A.K. Deodhar, R.E. Rana. Surgical physiology of wound healing: a review. J Postgrad Med, 43 (1997), pp. 52-56
5. W.V. Giannobile. Periodontal tissue engineering by growth factors. Bone, 19 (Suppl. 1) (1996), pp. 23S-37S
6. D.M. Dohan, J. Choukroun, A. Diss, *et al.* Platelet-rich fibrin (PRF): a second-generation platelet concentrate, part I: technological concept and evolution. Oral Surg Oral Med Oral Path Oral Radiol Endod, 101 (2006), pp. E37-E44

7. 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 U S A.* 1974;71:1207–1210. [PMC free article] [PubMed] [Google Scholar]
8. Kiran NK, Mukunda KS, Tilak Raj TN. Platelet concentrates: A promising innovation in dentistry. *J Dent Sci Res.* 2011;2:50–61.
9. Whitmann DH, Berry RL, Green DM. Platelet gel: an alternative to fibrin glue with applications in oral and maxillofacial surgery. *J Oral Maxillofac Surg.* 1997;55:1294–1299. [PubMed]
10. Choukroun J, Adda F, Schoeffler C, Vervelle A. Une opportunité en paro-implantologie: le PRF. *Implantodontie.* 2000;42:55–62. [Google Scholar]
11. Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, Gogly B. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part I: technological concepts and evolution. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e37–44. [PubMed]
12. Harrison P. Platelet function analysis. *Blood Rev.* 2005;19:111–123. [PubMed]
13. Castro HC, Ferreira BLA, Nagashima T, Schueler A, Rueff C, Camisasca D, Moreira G, Scovino G, Borges L, Leal M, Filgueira M, Paschoal P, Bernardo V, Bourguinhon S, Rodrigues CR, Santos DO. Plaquetas: ainda um alvo terapêutico. *J Bras Patol Med Lab.* 2006;42:321–332. [
14. Fernández-Delgado N, Hernández-Ramírez P, Forrellat-Barrios M. Platelet functional spectrum: from hemostasis to regenerative medicine. *Rev Cubana Hematol Inmunol Hemoter*. 2012;28:200–216.
15. Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, Gogly B. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part II: platelet-related biologic features. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e45–50.
16. Garcia VG, Corral I, Bascones-Martínez A. Plasma rico en plaquetas y su utilización en implantología dental. *Av Period.* 2004;16:81–92.
17. Gupta V, Bains BK, Singh GP, Mathur A, Bains R. Regenerative potential of platelet rich fibrin in dentistry: Literature review. *Asian J Oral Health Allied Sci.* 2011;1:22–28.
18. Gassling V, Douglas T, Warnke PH, Açil Y, Wiltfang J, Becker ST. Platelet-rich fibrin membranes as scaffolds for periosteal tissue engineering. *Clin Oral Implants Res.* 2010;21:543–549.
19. Laurens N, Koolwijk P, de Maat MP. Fibrin structure and wound healing. *J Thromb Haemost.* 2006;4:932–939. [PubMed]
20. Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, Gogly B. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part II: platelet-related biologic features. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e45–50. [PubMed]
21. Choukroun J, Diss A, Simonpieri A, Girard MO, Schoeffler C, Dohan SL, Dohan AJ, Mouhyi J, Dohan DM. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part IV: clinical effects on tissue healing. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e56–60. [PubMed]
22. Li Q, Pan S, Dangaria SJ, Gopinathan G, Kolokythas A, Chu S, Geng Y, Zhou Y, Luan X. Platelet-rich fibrin promotes periodontal regeneration and enhances alveolar bone augmentation. *Biomed Res Int.* 2013;2013:638043. [PMC free article] [PubMed]
23. Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, Gogly B. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part III: leucocyte activation: a new feature for platelet concentrates? *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e51–55. [PubMed]
24. Chang YC, Zhao JH. Effects of platelet-rich fibrin on human periodontal ligament fibroblasts and application for periodontal infrabony defects. *Aust Dent J.* 2011;56:365–371. [PubMed]
25. Kawase T, Kamiya M, Kobayashi M, Tanaka T, Okuda K, Wolff LF, Yoshie H. 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 B Appl Biomater.* 2015;103:825–31. [PubMed]

26. Wu CL, Lee SS, Tsai CH, Lu KH, Zhao JH, Chang YC. Platelet-rich fibrin increases cell attachment, proliferation and collagen-related protein expression of human osteoblasts. *Aust Dent J.* 2012;57:207–212. [PubMed]
27. 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;1:53–57. [PMC free article] [PubMed]
28. Bölükbaşı N, Ersanlı S, Keklikoğlu N, Başeğmez C, Ozdemir T. Sinus augmentation with platelet-rich fibrin in combination with bovine bone graft versus bovine bone graft in combination with collagen membrane. *J Oral Implantol.* 2013 [Epub ahead of print] [PubMed]
29. Joseph VR, Sam G, Amol NV. Clinical evaluation of autologous platelet rich fibrin in horizontal alveolar bony defects. *J Clin Diagn Res.* 2014;8:ZC43–47. [PMC free article] [PubMed]
30. Kim TH, Kim SH, Sándor GK, Kim YD. Comparison of platelet-rich plasma (PRP), platelet-rich fibrin (PRF), and concentrated growth factor (CGF) in rabbit-skull defect healing. *Arch Oral Biol.* 2014;59:550–558. [PubMed]
31. Simonpieri A, Del Corso M, Vervelle A, Jimbo R, Inchingolo F, Sammartino G, Dohan Ehrenfest DM. Current knowledge and perspectives for the use of platelet-rich plasma (PRP) and platelet-rich fibrin (PRF) in oral and maxillofacial surgery part 2: Bone graft, implant and reconstructive surgery. *Curr Pharm Biotechnol.* 2012;13:1231–1256. [PubMed]
32. Dohan Ehrenfest DM, Rasmusson L, Albrektsson T. Classification of platelet concentrates: From pure platelet-rich plasma (P-PRP) to leucocyte- and platelet-rich fibrin (L-PRF) *Trends Biotechnol.* 2009;27:158–67. [PubMed
33. Dohan Ehrenfest DM, Rasmusson L, Albrektsson T. Classification of platelet concentrates: From pure platelet-rich plasma (P-PRP) to leucocyte- and platelet-rich fibrin (L-PRF) *Trends Biotechnol.* 2009;27:158–67. [PubMed]
34. Anitua et al., Sanchez M, Orive G, Andia I. (2007): The potential impact of the preparation rich in growth factors (PRGF) in different medical fields. Biomaterials.28:4551–4560.
35. Dohan et al., Choukroun J, Diss A (2006c): Platelet-rich fibrin (PRF): A second-generation platelet concentrate. Part III: Leucocyte activation: A new feature for platelet concentrates? Oral Surgery, Oral Med Oral Pathol Oral Radiol Endodontology 101(3): e51-e55.
36. Dohan et al., Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J. (2006b): Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part II: platelet-related biologic features. Oral Surg Oral Med Oral Pathol Oral Radiol Endod.101:e45–e50.
37. Dohan et al., Del Corso M, Diss A, Mouhyi J, Charrier J-B. (2010): Three-Dimensional Architecture and Cell Composition of a Choukroun’s Platelet-Rich Fibrin Clot and Membrane. J Periodontol 81(4): 546-555.
38. Monitha G, Pavan B, Oza RR. Injectable Platelet-Rich Fibrin-A Revolution in Periodontal Regeneration. Cureus. 2022;14(8).
39. Pavlovic V, Ciric M, Jovanovic V, Trandafilovic M, Stojanovic P. Platelet-rich fibrin: Basics of biological actions and protocol modifications. Open Medicine. 2021 Mar 22;16(1):446-54.
40. Everts PA, Hoffmann J, Weibrich G, Mahoney CB, Schönberger JP, van Zundert A, et al. Differences in platelet growth factor release and leucocyte kinetics during autologous platelet gel formation. *Transfus Med.* 2006;16:363–8. [PubMed] [Google Scholar]
41. Dohan Ehrenfest DM, Del Corso M, Diss A, Mouhyi J, Charrier JB. Three-dimensional architecture and cell composition of a Choukroun's platelet-rich fibrin clot and membrane. *J Periodontol.* 2010;81:546–55. [PubMed] [Google Scholar]
42. Cromack DT, Porras-Reyes B, Mustoe TA. Current concepts in wound healing: Growth factor and macrophage interaction. *J Trauma.* 1990;30:S129–33. [PubMed] [Google Scholar]
43. Toffler M, Toscano N, Holtzclaw D, Corso MD, Dohan Ehrenfest DM. Introducing Choukroun's platelet rich fibrin (PRF) to the reconstructive surgery milieu. *J Implant Adv Clin Dent.* 2009;1:21–30. [Google Scholar]

44. Su CY, Kuo YP, Tseng YH, Su CH, Burnouf T. *In vitro* release of growth factors from platelet-rich fibrin (PRF): A proposal to optimize the clinical applications of PRF. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2009;108:56–61. [PubMed] [Google Scholar]
45. Dohan Ehrenfest DM, de Peppo GM, Doglioli P, Sammartino G. Slow release of growth factors and thrombospondin-1 in Choukroun's platelet-rich fibrin (PRF): A gold standard to achieve for all surgical platelet concentrates technologies. *Growth Factors.* 2009;27:63–9. [PubMed] [Google Scholar]
46. Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, et al. Platelet-rich fibrin (PRF): A second-generation platelet concentrate. Part II: Platelet-related biologic features. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e45–50. [PubMed] [Google Scholar]
47. Choukroun J, Diss A, Simonpieri A, Girard MO, Schoeffler C, Dohan SL, et al. Platelet-rich fibrin (PRF): A second-generation platelet concentrate. Part IV: Clinical effects on tissue healing. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:e56–60. [PubMed]
48. Verma UP, Yadav RK, Dixit M, Gupta A. Platelet-rich fibrin: a paradigm in periodontal therapy–a systematic review. Journal of international society of preventive & community dentistry. 2017 Sep;7(5):227.
49. Chang YC, Zhao JH. Effects of platelet-rich fibrin on human periodontal ligament fibroblasts and application for periodontal infrabony defects. *Aust Dent J.* 2011;56:365–371. [PubMed]
50. Molly L, Quirynen M, Michiels K, Van Steenberghe D. Comparison between jaw bone augmentation by means of a stiff occlusive titanium membrane or an autologous hip graft: a retrospective clinical assessment. *Clin Oral Implants Res.* 2006;17:481–487.
51. Simonpieri A, Del Corso M, Vervelle A, Jimbo R, Inchingolo F, Sammartino G, Dohan Ehrenfest DM. Current knowledge and perspectives for the use of platelet-rich plasma (PRP) and platelet-rich fibrin (PRF) in oral and maxillofacial surgery part 2: Bone graft, implant and reconstructive surgery. *Curr Pharm Biotechnol.* 2012;13:1231–1256. [PubMed]
52. Del Corso M, Toffler M, Dohan Ehrenfest DM. Use of an autologous leukocyte and platelet-rich fibrin (L-PRF) membrane in post-avulsion sites: an overview of Choukroun's PRF. *J Implant Adv Clin Dent.* 2010;1:27–35.
53. Simonpieri A, Choukroun J, Del Corso M, Sammartino G, Dohan Ehrenfest DM. Simultaneous sinus-lift and implantation using microthreaded implants and leukocyte- and platelet-rich fibrin as sole grafting material: a six-year experience. *Implant Dent.* 2011;20:2–12. [PubMed]
54. Mazor Z, Horowitz RA, Del Corso M, Prasad HS, Rohrer MD, Dohan Ehrenfest DM. Sinus floor augmentation with simultaneous implant placement using Choukroun's platelet-rich fibrin as the sole grafting material: a radiologic and histologic study at 6 months. *J Periodontol.* 2009;80:2056–2064. [PubMed]
55. Toffler M, Toscano N, Holtzclaw D, Corso MD, Dohan Ehrenfest DM. Introducing Choukroun's platelet rich fibrin (PRF) to the reconstructive surgery milieu. *J Implant Clin Adv Dent.* 2009;1:21–30.
56. Choukroun J, Diss A, Simonpieri A, Girard MO, Schoeffler C, Dohan SL, Dohan AJ, Mouhyi J, Dohan DM. Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part V: histologic evaluations of PRF effects on bone allograft maturation in sinus lift. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.* 2006;101:299–303. [PubMed]
57. Yilmaz D, Dogan N, Ozkan A, Sencimen M, Ora BE, Mutlu I. Effect of platelet rich fibrin and beta tricalcium phosphate on bone healing. A histological study in pigs. *Acta Cir Bras.* 2014;29:59–65. [PubMed]