A comprehensive clinical review of Platelet Rich Fibrin (PRF) and its role in promoting tissue healing and regeneration in dentistry. Part II: Preparation, optimization, handling and application, benefits and limitations of PRF

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Summary
The purpose of Part II of this review is to analyse the available literature on PRF relating to: (i) preparation technique; (ii) optimizing the quality of PRF; (iii) its physical handling and application; and (iv) its benefits and limitations. An improved understanding of these aspects will facilitate the clinicians’ ability to enhance the therapeutic applications of PRF in the fields of dental implantology, periodontology and oral surgery.

PRF, a patient blood-derived living biomaterial, is increasingly being investigated and used worldwide by clinicians as an adjunctive autologous biomaterial to promote bone and soft tissue healing and regeneration. PRF technology has grabbed the attention of clinicians because this biomaterial is derived from the patients’ own blood; is readily available; easy to prepare; can be produced immediately at chair-side; easy to use; and widely applicable in dentistry, whilst being financially realistic for the patient and the clinician, and with virtually no risk of a rejection reaction (foreign body response). The 3D architecture of the fibrin matrix provides the PRF membrane with great density, elasticity, flexibility and strength that are excellently suited for handling, manipulation and suturing. Optimal PRF membrane quality and treatment success is dependent on: quick collection of blood and transfer to the centrifuge (2 minutes as per Choukroun); use of proper centrifugation protocol; maturation of the clot for 4-8 minutes before use; preparation of the membrane using a standardized preparation technique; and appropriate conservation of the membrane before use. The PRF can be used as a membrane (A-PRF or L-PRF), liquid or injectable form (i-PRF), plug or the membrane can be cut in fragments, and applied either in stand-alone therapies (i.e. plug, filler or protective barrier); additive therapies (i.e. added or mixed to bone substitutes); or used in combination therapies with other biomaterials (i.e. protective barrier in GBR procedures).

At present, very little is understood about PRF generated from patients with coagulation disorders or patients on medications that affect blood clotting (heparin, warfarin or platelet inhibitors). Its lack of rigidity and fast degradation (biodegradability) may limit its application as a sole barrier membrane in GTR procedures. One of the clinical limitations to deal with is the heterogeneity in the quality of platelets and blood components of various PRF protocols. At this stage in time there is not a single RCT or CCT to compare the effectiveness of A-PRF or L-PRF protocols. Furthermore, in vitro studies that claim superiority or inferiority of a specific PRF preparation have yet to be validated by independent clinical trials. Standardized and optimized PRF preparation protocols, and their effectiveness still have to be validated through independent robust randomized controlled clinical trials.

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Introduction

The prospect of having new therapies, biomaterials and bioactive surgical additives available that will improve success and predictability of patient outcomes in soft and bone tissue healing and regeneration are key treatment objectives in dental implantology, periodontology and oral surgery.

Platelet Rich Fibrin (PRF), a patient blood-derived and autogenous living biomaterial, is increasingly being investigated and used worldwide by clinicians as an adjunctive autologous biomaterial to promote bone and soft tissue healing and regeneration. The gold standard for in vivo tissue healing and regeneration requires the mutual interaction between a scaffold (fibrin matrix), platelets, growth factors, leukocytes, and stem cells. These key elements are all active components of PRF, and when combined and prepared properly are involved in the key processes of tissue healing and regeneration, including cell proliferation and differentiation, extracellular matrix synthesis, chemotaxis and angiogenesis neo-vascularization. An improved understanding of the development, biological and physiological properties and characteristics of PRF in tissue healing and regeneration over the last two decades, has led to more successful therapeutic applications, especially in the fields of dental implantology, periodontology and oral surgery.

The purpose of this comprehensive review is to analyse the available scientific literature on PRF regarding its: (Part I) (i) definition and purpose in the clinical environment; (ii) development and classification of platelet concentrate biomaterials; (iii) biological characteristics and composition; (Part II) (iv) preparation technique; (v) optimizing quality; (vi) physical application and handling; and (vii) the benefits and limitations of PRF; and (Part III) (viii) its clinical applications in implant dentistry, periodontics, oral surgery and regenerative endodontics.

Methodology, Search strategy and inclusion criteria

An electronic MEDLINE (PubMed) and Google Scholar search was performed for all articles on Platelet Rich Fibrin (PRF) and Platelet concentrates up to May 2016. The search was complimented by an additional hand search of selected journals in oral implantology, oral surgery and periodontal, as well as grey literature. The reference lists and bibliographies of all included publications were also screened for relevant studies. The search was limited to the English language. Randomized controlled trials (RCT’s), controlled clinical trials (CCT’s), case reports, case series, prospective, retrospective and in-vitro/in vivo studies were included in the narrative review. Animal studies were excluded from this review.

How is PRF prepared and how can I use it?

Blood drawing

A major advantage of PRF is that it has a simple preparation protocol. Blood is drawn from the patient using a sterile 10mL vacutainer (2-12 tubes) just before or during surgery. (Figure 1.) The dental clinician has the following options for drawing blood; (i) doing it himself (venepuncture courses are available); (ii) anaesthetist or sedationist when using general anaesthesia or conscious sedation for procedures; or (iii) contracting the services of a qualified nurse.
Centrifugation

The tubes should always be balanced by opposing two tubes to equilibrate the centrifugation forces and to prevent vibrations during the centrifugation process. (Figure 2) At the end of the centrifugation spin, the A-PRF or L-PRF (not i-PRF) caps are removed and the tubes placed in a sterile tube holder. (Figure 3) The blood sample with clot is allowed to rest/mature for approximately 4-8 minutes before extracting the clot from the tube. (Figure 4)

The centrifugation process activates the coagulation process and separates the blood sample into three different layers: an acellular plasma at the top of the tube; a strongly polymerized fibrin clot is formed in the middle; and blood cells (red corpuscle base) are gathered at the bottom of the tube.4,5,6

Currently used centrifugation protocols

There are various centrifugation processing-protocols that are currently being used.

- Original Choukroun’s PRF protocol (standard protocol): 3000 rpm / 10 minutes
- Dohan Ehrenfest’s Group - Leukocyte- and Platelet-Rich Fibrin (L-PRF): Speed 2700 rpm / 12 minutes
- Choukroun’s advanced PRF (A-PRF), enriched with leukocytes: 1300 rpm / 8 minutes
- Choukroun’s i-PRF (solution/gel): 700rpm/3 minutes

Effect of centrifugation protocols on the optimum fibrin clot: cell ratio

Current data show that there is a differential distribution of red blood cells, platelets, and leukocytes in the PRF clot.
Raton, FL, USA) and covered with the lid. The PRF membranes are ready for use after 2 minutes. It provides a 3-dimensional matrix or scaffold that contains high concentrations of platelets, leukocytes and growth factors. A PRF membrane remains usable many hours after preparation, as long as the PRF is prepared correctly and conserved in physiologic conditions. Moreover, the use of the PRF Box is a user-friendly and inexpensive tool, allows for standardized preparation of homogeneous PRF membranes with a higher growth factor content, avoids the dehydration or death of the leukocytes living in the PRF clot, and also prevents the shrinkage of the fibrin matrix architecture.12,13,14

The concept of ‘the optimal PRF scaffold or composites’, tailored for specific clinical applications, is still in a process of virtual development. Further clinical trials are required to validate this concept, and what centrifugal force and time is required to get the optimum PRF composite biomaterial for specific clinical applications.

At this stage in time there is not a single RCT or CCT to compare the effectiveness of any of the above-mentioned PRF (A-PRF or L-PRF) protocols. Furthermore, in vitro studies that claim superiority or inferiority of a specific PRF preparation have not been validated by independent clinical trials. Therefore, based on these facts, no preference or distinction is made between any of the above-mentioned PRF preparation protocols (A-PRF or L-PRF) in this review.

Effect of the type of test tube and compression on the clot quality
Research data suggest that the type of vacutube that is used (i.e. dry glass or glass-coated plastic tubes) and the compression process of the clot (forcible or soft) do not seem to influence the architecture of this autologous biomaterial. However, both parameters could influence the growth-factor content and the matrix properties of the product.5 The influence of these preparation factors requires further analysis and there effect on the clinical efficacy needs to be validated with good quality clinical trials.

Preparation of PRF membranes
Each fibrin clot concentrates most platelets (97%) and more than half of the leukocytes from a 9-ml blood harvest.6,10,11 The PRF clot is removed from the tube with a sterile tweezer. The fibrin clot is separated from the red blood cell fragment, approximately 2 mm below the dividing line, using a scissor. (Figure 5) The section of the blood clot attached to the fibrin clot contains the stem cells. The PRF clots are placed in the Box grid (Process, France) or Xpression Kit (Intra-Lock, Boca-

Figure 5: The fibrin clot is separated from the red blood cell fragment using a scissor.
Practical guidelines for optimizing the quality of PRF

PRF is a living biomaterial that requires a good knowledge and skills on how to produce, prepare, conserve and use it most effectively and efficiently.14,6,15 Incorrect use could lead to damaged dry product leading to inconsistent clinical results.16 The most critical factor affecting the success of PRF in healing and regenerative therapy is the quality of PRF preparations. Following are some practical guidelines how to optimize the quality of PRF, increase clinical efficiency and consistency, and how to prevent common mistakes.

- **Limit centrifuge vibration during PRF preparation**
  Centrifuge rotational speed and subsequent vibrations, has been shown to have a directly impact on the architecture and cell content of a PRF clot.17 The Dohan group have hypothesized that the type/make of centrifuge machine has an effect on rotational speed and vibrations. The latter however, has not been validated with clinical trials.
  Vibrations can be limited by following the following rules:
  (i) Always make sure that the centrifuge tubes are filled equally (1 cm from the top); (ii) Always balance the rotor properly. Every tube must have a balance or opposing tube (Figure 2). (iii) Do not balance with a vacutube filled with water, the distribution of the densities will be incorrect and cause unnecessary vibration; (iv) If properly balanced and used, the rotor should accelerate smoothly and with a constant change in the pitch of the motor sound. (v) Any vibrations, or unusual sounds should cause the cessation of operation immediately by the operator; (vi) Initial vibrations with start of centrifugation can be reduced by holding your hand on the lid; (vii) Never leave the centrifuge until you are certain that it has reached its operating speed and is functioning properly. All rotors go through a minor vibration phase when they first start. There will be a minor flutter when the rotor reaches this vibration point - do not confuse this with a serious vibration caused by imbalance.

- **Patients receiving anticoagulants**
  At present, very little is understood about PRF generated from patients with coagulation disorders or patients on medications that affect blood clotting (heparin, warfarin or platelet inhibitors). When patients are on any type of anticoagulant therapy, they have a longer coagulation time, therefore it is suggested to centrifuge blood for longer periods, or increase the waiting time after centrifugation (approxim ately 5-10 minutes). However, there is no data available to support this recommendation.

- **Standardized and efficient preparation of PRF**
  The PRF box (Box grid, Process, Nice, France or Xpression Kit, Intra-Lock, Boca-Raton, FL, USA) was designed to collect and transform up to 16 PRF clots into membranes in sterile conditions and to conserve them in a clean and wet environment before use. (Figure 6a) The box also contains compression wells and maces to compress the PRF clots into dense PRF cylinders, easy to use for filling cavities (such as extraction sockets). It is suggested that gentle pressure is used to prevent squeezing out all of the plasma contained in the original PRF clots.18 Compressing the clot too hard or too long results in shrinkage of the fibrin network, release of growth factors, dehydration and damage of leukocyte content.13 PRF membranes or plugs are ready for use within 2 minutes after compression of clots in the Box grid (Figure 6b) or plug well (cylinder).

  The serum exudate collected in the bottom of the box, can be used for a longer conservation of the membranes, and is ready to be mixed with a bone biomaterial for grafting. This standardized approach also allows an increase in the total growth factors release of the PRF membrane itself.13
part of the clot contains the highest concentration of platelets and stem cells required for bone regeneration. Thus, it is necessary to preserve a small RBC layer at the PRF clot end to collect as many platelets and leukocytes as possible. This part of the procedure is done with scissors and remains operator-dependent.

• Optimizing and preserving growth factor release

For the standardization of PRF preparations as a grafting material for tissue regeneration, PRF membranes should always be preserved in a wet serum environment. The compression procedure of the clots into membranes is performed with a gentle, slow, and homogeneous pressure to prevent squeezing out all the serum contained in the original PRF clot. This will ensure that the final membrane always remains homogeneously wet and soaked with serum. This gentle method thus avoids extracting and losing a significant amount of extrinsic-incorporated platelet growth factors.

Significant amounts of growth factors are released during the first 20 minutes after preparation. PRF membranes should therefore be used as quickly as possible after preparation. However, this release is considerably less significant when forcible extraction is avoided. In physiologic conditions, the main release occurs after several hours, and PRF can be used a long time after preparation, as long as the material is conserved in the adequate conditions.

• Handling the PRF (Clinical application)

PRF membranes are easy to drape over a surgical or augmented site. The elastic consistency of the PRF membrane also allows the clinician to punch a hole in the membrane to drape over a healing abutment before suturing the flap.

The exudate in the bottom of the box is rich in proteins (Vitronectin and Fibronectin). This solution can be recovered with a syringe and used to hydrate biomaterials, flush the surgical site, wet the implant surface and to preserve harvested autogenous bone blocks, rather than using saline.

• Conservation of the PRF membrane

PRF is a blood-derived living tissue and must be handled carefully to keep its cellular content alive and stable. Conserve the PRF clot in its centrifugation tube. As long as the serum has not been flushed away from the clot, the growth factor content remains stable. It is a good way to gain 5-15 minutes.

PRF membranes remain usable many hours after preparation, as long as the PRF is prepared correctly and conserved in physiologic conditions.

The PRF Box, a user-friendly and inexpensive tool, allows to guarantee the adequate preparation of homogeneous PRF membranes with a higher growth factor content, avoids dehydration or death of the leukocytes living in the PRF clot, and also prevents shrinkage of the fibrin matrix architecture.

• Optimal preparation and selection

For the standardization of PRF preparations and to preserve platelets and growth factors it is suggested not to squeeze out all of the plasma contained in the original PRF clots. Platelets are not equally distributed inside and on the surface of the PRF clot. Therefore, in a clinical situation, when growth factors provided by platelets are expected and desired, the platelet-rich region adjacent to the red thrombus should be used. Therefore, always place the part of the clot closest to the thrombus closest to the grafting site. This part of the clot contains the highest concentration of platelets and stem cells required for bone regeneration. Thus, it is necessary to preserve a small RBC layer at the PRF clot end to collect as many platelets and leukocytes as possible. This part of the procedure is done with scissors and remains operator-dependent.
How can I use PRF? (Physical application)

PRF (A-PRF or L-PRF) can either be used as a clot (Figure 10a), membrane (Figure 10b), injectable liquid (i-PRF) (Figure 10c), plug (Figure 10d), or the membrane can be cut up in fragments (Figure 10e & 10f). PRF can either be applied in stand-alone, additive, or in combination therapies.

• Stand-alone therapies

Typical stand-alone therapies include using the fibrin plug or membrane as a filler material in extraction sockets (Figure 7a & 7b) Mixing autogenous bone or bone substitutes (allografts) with i-PRF (PRF Liquid) for use in GBR procedures transforms particulate bone into a easily to handle gel consistency (Figure 8a & 8b).

PRF liquid (i-PRF) can be injected above, or PRF (A-PRF or L-PRF) membrane placed above the GBR or GTR membrane (Figure 9a & 9b) to act as a interposition barrier to protect and stimulate the bone compartment, and as a healing membrane in order to improve the soft tissue healing and remodeling, and thus avoid soft tissue dehiscence. 21,34

Figure 8a: i-PRF added to particulate bone.

Figure 8b: i-PRF transforms the graft material into an easy to handle gel consistency

Figure 9a: Collagen membrane placed over bone graft.

Figure 9b: PRF membrane placed above a GBR membrane as interposition barrier to promote soft tissue healing.
**Combination therapies**

The PRF membrane is typically used in combination with other biomaterials in bone augmentation and grafting sites as a graft material or barrier membrane. The purpose of PRF is to activate and facilitate the healing and regenerative capacity of the host tissue, by providing a strong fibrin scaffold, major growth factors and allowing space for tissue regeneration. Using PRF as a protective barriers on bone graft sites helps to avoid perforations of the weakened gingival tissues and to prevent associated contamination of the bone graft below.

**Key biological function of a PRF membrane**

PRF membranes are not comparable to heterologous resorbable collagen or non-resorbable (i.e. dPTFE) membranes. PRF membranes belong to a completely different category of membrane, namely natural autologous...
(iii) Protective barrier and healing booster

PRF membranes are frequently used for the protection of the grafted area and as a healing booster for the soft tissues above the grafted defects or augmented sites. The purpose of the PRF membrane is not only to protect the blood clot and/or the graft material, like in the GTR concept, but also to promote the induction of a strong and thick periosteum and gingiva. This boosted periosteum functions as a true barrier between the soft tissue and bone compartments, and constitutes probably the best protection and regenerative barrier for the intrabony defects.

What are the benefits of PRF?

PRF has increasingly grabbed the attention of clinicians worldwide because this biomaterial is of natural origin (autologous / derived from the patients’ own blood), can be produced chair side, is easy to make, readily available, easy to use within the daily clinical routine; widely applicable in dentistry, whilst being financially realistic for the patient and the health care system.

The following major advantages support the clinical use of PRF in tissue repair and regeneration processes.

- Natural (autologous) biomaterial

Regenerative therapies are now shifting away from using allogenic and xenogenic biomaterials to autologous biomaterials. While other membranes are considered as foreign bodies by the host tissues and interfere with the natural tissue healing process, a PRF membrane is as natural as the host tissue with virtually no risk of infection, immune or a rejection reaction (foreign body response).
• **Easy and efficient to use**

Preparing PRF is easy, fast and user-friendly to use within the daily clinical routine.\(^9,19,39,40,41\) PRF is prepared on site (in the clinical setting) simply by drawing blood from the patient for immediate use thus reducing patient waiting time. Currently used protocols for the preparation of autologous PRF are standardized and easy for clinicians and clinical assistants to use. Furthermore, there is no limitation on quantity of PRF membranes required.

In comparison with a natural blood clot, a PRF membrane is a solid material, has a strong fibrin architecture, and easier to handle and to position.\(^9,38\)

Fibrin 3D architecture provides the PRF membrane with great density, elasticity, flexibility and strength,\(^4,40\) that is better suited for manipulation and suturing.\(^7\) The elastic consistency of the PRF membrane allows the clinician to punch it to allow for placing it around a healing or prosthetic abutment\(^40\) or even suture it.

• **Increased clinical performance**

Recent studies have shown that, compared with previous generation PRP, PRF exhibits a greater expression and concentration of growth factors and matrix proteins, which are released more slowly due to the three-dimensional architecture of the adhesive glycoproteins in the fibrin, which results in significantly better performance.\(^12,13,42\)

Moreover, A-PRF shows a more gradual release of growth factors, up to a 10-day period, and stimulated significantly higher growth factor release over time when compared to Choukroun’s standard PRF.\(^9\)

• **Safety and Low risk**

Blood is drawn from the patient and therefore reduced donor site morbidity. PRF rarely causes complications such as membrane exposure, an unwanted outcome that has been observed in cases using biodegradable barrier membranes.\(^43\) A further advantage of PRF is the extremely low risk of infection. Moreover, no in vitro cytotoxicity effects were detected whatever the quantity of PRF used.\(^44,45,46\)

• **Increased healing potential**

PRF increases the predictability of wound healing and regeneration potential of tissues.\(^24,47,48,49,50,51,52,53\)

• **Reduced morbidity**

A clinical advantage of PRF as a graft material is related to avoidance of a donor site and risk of morbidity, thus resulting in a decrease in patient discomfort, postsurgical pain, and bleeding after operation.\(^20,54\) PRF is not only a platelet concentrate but also an ‘immune node’ that is able to stimulate defence mechanisms. It is even likely that the significant inflammatory regulation noted on surgical sites treated with PRF, is the outcome of retro-control effects from cytokines trapped in the fibrin network and released during the remodeling of this initial matrix.\(^11\) Furthermore, evidence suggest that the content of platelet alpha granules might have

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**Figure 13:** PRF membrane fragments mixed with bone particulate.

**Figure 14:** PRF membrane used as a protective and regenerative barrier for intrabony defect.
a bactericidal effect, mediated by molecules called thrombocidines that may have an important contribution toward reducing post-operative infections. The antimicrobial properties of PRF are also advantageous and convenient during surgical procedures.

**Cost-benefit**

PRF has a potential outstanding cost to benefit ratio. A platelet-rich fibrin (PRF) membrane is a readily available and inexpensive biomaterial that is beneficial in implant dentistry, oral surgery and periodontal procedures.

The ease of preparation and cost-effectiveness of PRF membrane offers a huge advantage over other commercially available membranes. It is widely applicable in dentistry, whilst being financially realistic for the patient and the health care system. PRF is currently the safest and most economical choice for patients and clinicians for improving healing and tissue (soft tissue and bone) regeneration outcomes. It can spare the patient from additional operating field (second surgery mobility) and also save cost through avoiding use of alloplastic or xenogenic membranes and reduce the quantity of synthetic grafting materials. It is also an economical alternative to expensive recombinant growth factors when used in conjunction with osseous grafts.

**Affordability and cost effectiveness**

PRF is a simple and inexpensive biomaterial to use. The initial investment for producing PRF preparations, which includes centrifuge, instrument and box kit, currently does not exceed ZAR 28,000 (US$ 1806) for the A-PRF Duo™ system and approximately US$ 3950 for the Intra-Spin™ (Intra-Lock) L-PRF system (prices as quoted in June, 2016, excluding shipping and courier costs). Therefore, it would be potentially affordable for most private practices to invest long-term in basic equipment to produce PRF preparations.

**No contraindications**

PRF membranes have no contraindications, they can be used in all kinds of patients, especially in patients with systemic conditions where healing is compromised (i.e. diabetics and smokers), or in surgically compromised situations (damaged flap). In these situations PRF will promote soft tissue healing and stimulate the healing of a damaged flap and reduce the risks of flap necrosis after a surgery. It is a common point that all fibrin-based products (platelet concentrates), are frequently used for the stimulation of angiogenesis, and to reduce the risk of flap necrosis in many general surgery applications.

**Open Access and widely applicable**

The PRF technique is open-access and thus can be widely developed and used in private practice without commercial considerations. The clinical applications of PRF in other fields of dentistry, i.e. endodontics are increasing exponentially.

**Limitations**

The rapid use of the PRF without delay or short handling time may be a potential limitation. Lack of rigidity and fast degradation (biodegradability) may limit its application in GTR procedures. PRF can be considered a healing biomaterial that can be utilized in regenerative surgical procedures to fasten healing, but its application as a barrier membrane in GBR is doubtful due to its poor mechanical properties. Owing to the fact that PRF is an autologous product, the availability of this biomaterial in larger amounts is also a concern. One of the clinical limitations to deal with is the heterogeneity in the quality of platelets and blood components. At present, very little is understood about PRF generated from patients with coagulation disorders or patients on medications that affect blood clotting (heparin, warfarin or platelet inhibitors).

**Conclusion**

The therapeutic use of PRF for accelerating tissue healing and regeneration has increasingly grabbed the attention of clinicians world-wide because this biomaterial is of natural origin (autologous / derived from the patients’ own blood); PRF technology is readily available; is easy to prepare; can be produced immediately at chair-side; easy to use; and widely applicable in dentistry, whilst being financially realistic for the patient and the clinician, and with virtually no risk of a rejection reaction (foreign body response).

The 3D architecture of fibrin provides the PRF membrane with great density, elasticity, flexibility and strength that are excellently suited for handling, manipulation and suturing. Optimal PRF membrane quality and treatment success is dependent on: quick collection of blood and transfer to the centrifuge; use of proper centrifugation protocol; maturation of the clot for 5 minutes before use; preparation of the membrane using a standardized preparation technique; and appropriate conservation of the membrane before use.

The PRF can be used as a membrane (A-PRF or L-PRF), gel (i-PRF), plug or the membrane can be cut in fragments, and applied either in stand-alone therapies (i.e. plug, filler or protective barrier), additive therapies (i.e. added or mixed to bone substitutes); or used in combination therapies with...
other biomaterials (i.e. protective barrier in GBR procedures). More importantly, the use of PRF enables local delivery of a fibrin matrix, cells, growth factors and proteins that provide unique biological properties and cues for promoting new blood vessel formation, and potentially accelerating wound healing and tissue regeneration, whilst at the same time reducing adverse events. Consequently, the benefits of PRF in wound and bone healing, its antibacterial and antithrombotic effects, the low risks with its use, and the availability of easy and low cost preparation methods should encourage more clinicians to adopt this technology in their practice for the benefit of their patients.

One of the clinical limitations to deal with is the heterogeneity in the quality and quantity of platelets and blood components due to use of different PRF preparation protocols. At this stage in time there is not a single RCT or CCT to compare the effectiveness of any of the PRF (A-PRF or L-PRF) protocols. Furthermore, in vitro studies that claim superiority of inferiority of a specific PRF preparation have not been validated by independent clinical trials. PRF preparation protocols (A-PRF and L-PRF) and the clinical effectiveness thereof still have to be validated and standardized through independent randomized controlled clinical trials.

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