Received: 19 September 2018 | Revised: 14 January 2019 | Accepted: 20 January 2019

DOI: 10.1002/JPER.18-0553

JOURNAL OF Periodontology

GUEST EDITORIAL

Standardization of relative centrifugal forces in studies related to platelet-rich fibrin

Richard J. Miron[1,2*] | Nelson R. Pinto[3,4*] | Marc Quirynen[4] | Shahram Ghanaati[5]

[1]Department of Periodontology, University of Bern, Bern, Switzerland

[2]Post-Doctoral Periodontology Program, Laboratory for Immunoregulation and Tissue Engineering (LITE), Division of Periodontology, Diagnostic Sciences & Dental Hygiene, University of Southern California, Ostrow School of Dentistry, Los Angeles, CA, USA

[3]Department of Periodontology and Implant Dentistry, University of the Andes (UANDES), Santiago, Chile

[4]Department of Oral Health Sciences, Katholieke Universiteit Leuven (KUL), Department of Periodontology, University Hospitals Leuven, Leuven, Belgium

[5]FORM, Frankfurt Oral Regenerative Medicine, Clinic for Maxillofacial and Plastic Surgery, Johann Wolfgang Goethe University, Frankfurt Am Main, Germany

Correspondence
Richard J. Miron, Department of Periodontology, University of Bern, Bern, Switzerland.
Email: richard.miron@zmk.unibe.ch

*Richard J. Miron and Nelson R. Pinto contributed equally to this work.

Abstract

Platelet-rich fibrin (PRF), a second-generation platelet concentrate, has been the focus of intensive research endeavors over the past 2 decades. Over the years, however, numerous reports have inaccurately reported relative centrifugal force (RCF) values, which has caused considerable confusion in the field. Furthermore, the use of trade names such as leukocyte and platelet-rich fibrin (L-PRF) and advanced platelet-rich fibrin (A-PRF) has further confused many readers, since studies have not always used centrifugation parameters with equal rotor sizes, angulation of tubes, and/or tube design. This has led to considerable misperception in the report of relative centrifugal force. Herein is described necessary parameters pivotal for the future report of RCF in studies related to PRF, which include: 1) dimensions of the rotor (radius at the clot and end of the tube); 2) rotor angulation for the tube holder; 3) revolutions per minute (RPM) and time; 4) RCF value calculated at either the RCF-minimum, RCF-clot, or RCF-maximum; 5) composition and size of tubes used to produce PRF; and 6) centrifugation model used. This editorial aims to minimize confusion in the field and create more transparent research reporting RCF values in future studies.

KEYWORDS

advanced platelet-rich fibrin, A-PRF, leukocyte and platelet-rich fibrin, L-PRF, platelet-rich fibrin, PRF

1 | INTRODUCTION

Platelet-rich fibrin (PRF) has been extensively studied as a regenerative material capable of facilitating hard and soft tissue regeneration under various clinical indications.[1–4] Various research groups have further shown that different biological properties of PRF collected from a given individual may result based on centrifugation speeds; i.e. relative centrifugal forces (RCFs).[5–9] Over the years however, numerous reports have misrepresented RCF values which have been re-transcribed in a several studies by various authors.[10] Various research groups have continued to misrepresent RCF values with little detail provided on the rotor sizes, rotor angulation, tube sizes and/or manufacturer of the centrifugation system and this article aims to define a guideline for future studies reporting RCF values in studies related to PRF.[11] One of the confusions that has been created in the field over the years is that various research groups may report RCF values at the PRF clot (referred to as RCF-clot; location at which the PRF clot is formed),[12–14] whereas others have reported RCF values either at the minimum[15,16] or maximum[9,17–20] of centrifugation tubes (referred to as RCF-min or RCF-max). This article aims to clarify these misunderstandings and proposes that a consensus be reached regarding more accurate means to report g-force values in future studies investigating PRF.

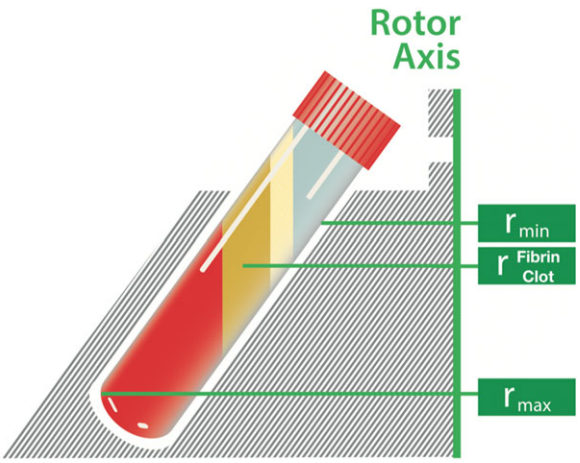

FIGURE 1 Graphic representation of a centrifugation with the standard locations where the "relative centrifugal force" (RCF) or gravity (g) is measured. Both depend on several parameters, including: the speed (rotation/revolutions per minute, RPM), the rotor diameter and rotor angulation. Standard locations for the calculation of RCF for a centrifuge with a rotor with fixed angle are: the minimal r_{min} at the top inside (shortest distance to rotor), the clot r_{clot} in the middle of the fibrin clot (middle of the tube), and maximal RCF value r_{max} (at largest distance to rotor)

2 | DEFINITION AND CALCULATION OF RELATIVE CENTRIFUGAL FORCE

One of the areas that has led to considerable confusion over the years is that RCF values have been calculated at various regions along a centrifugation tube without reporting where these RCF values were derived. Since RCF values are subject to significant changes depending on the rotor radius (distance between the tube and the rotor axis) (Fig. 1), it is important to understand that an increase in radius simply caused by changes in rotor angulations and rotor diameter has a dramatic effect on RCF values. For these reasons, it is important to have a basic understanding of RCF values including calculations to obtain RCF-min, RCF-clot, and RCF-max. The formula for RCF ($RCF = 11.18 \times r \times (N/1{,}000)^2$ where N is revolutions per minute and r is the radius in cm)[21] favors greater values at larger radiuses since the radius plays a multiplying role on final RCF values. As depicted in Figure 1 representing a centrifuge (which typically range in angulation from 30° to 45°), RCF values can easily be doubled between the RCF-min and RCF-max based on this increased radius distance (Table 1). Another confusion that has been created and expressed in several articles is that many studies related to PRF have often used RCF-min, RCF-clot, or RCF-max values without reporting exactly where these RCF values were derived. This has caused significant difficulty for researchers to further reproduce data, and a general lack of understanding has been created as a result. Herein we propose that all future articles report the following parameters in studies related to PRF: 1) dimensions of the rotor (radius at the clot and end of the tube); 2) rotor angulation for the tube holder; 3) revolutions per minute (RPM) and time; 4) RCF value calculated at either the RCF-min, RCF-clot, or RCF-max; 5) composition and size of tubes used to produce PRF; and 6) centrifugation model used.

TABLE 1 RCF calculation (minimum: RCF_{min}, average: RCF_{av}, maximum: RCF_{max}, and RCF_{clot} where clot is formed, respectively) using the calculator for the two most frequently used centrifuges for the preparation of PRF: the IntraSpin L-PRF centrifuge and the Duo Centrifuge, depending on the rotor angulation and RPM

Device	RCF_{min}	RCF_{av}	RCF_{max}	RCF_{clot}
IntraSpin				
Rotor angulation: 33°				
Distance to rotor in mm	40 mm	60 mm	80 mm	50 mm
2,700 RPM	326	489	653	408
PRF Duo				
Rotor angulation: 40°				
Distance to rotor in mm	63 mm	87 mm	110 mm	77 mm
1,300 RPM	119	164	208	145
2,700 RPM	513	708	897	628

RPM = rotations per minute; RCF = relative centrifugal force.
If the RPM and rotor radius are known, a nomogram chart[22] (Rickwood, p. 287) can be used to determine the RCF. RCF (or g) can be calculated by using the following formula: $RCF = 11.18 \times r \times (N/1{,}000)^2$
For this equation, r is the radius in centimeters from the center of the rotor to the sample during centrifugation and N is the rotor speed in RPM.

Each of the above-mentioned parameters may influence regeneration success with PRF. It is therefore appropriate to emphasize the importance of both standardizing and reporting of RCF parameters as a major factor under the control of the clinician. Only when the factors under the clinician's control are standardized can both researchers and clinicians begin to assess other variables that may influence clinical outcomes to the extent that they should be standardized.

An example of text to include in a Materials & Methods section would present as follows: "Leukocyte and Platelet-Rich Fibrin (L-PRF) membranes were produced using a protocol of 2,700 RPM for 12 minutes (RCF-clot = 408 g). L-PRF membranes were produced using an Intraspin centrifugation device (33° rotor angulation, 50 mm radius at the clot, 80 mm at the maximum, Intra-Lock, Boca Raton, Florida) using 9-mL glass-coated plastic tubes (Intra-Lock)."

Similarly, a second example using A-PRF may be described as follows: "Advanced Platelet-Rich Fibrin plus (A-PRF+) membranes were produced using a protocol of 1,300 RPM for 8 minutes (RCF-max = 208 g). A-PRF+ membranes were produced with 10-mL glass tubes using a Duo Quattro centrifugation device with a 40° rotor angulation with a radius of 88 mm at the clot and 110 mm at the max (Process for PRF, Nice, France)."

In this way, additional parameters are included in each of the Materials & Methods sections of future publications

on the topic with a more accurate ability to reproduce the data set.

3 | USE OF TRADE NAMES IN STUDIES RELATED TO PLATELET-RICH FIBRIN

One of the disadvantages in the literature has been the overlapping use of various trade names such as L-PRF in a variety of studies that have not appropriately used proprietary protocols. As previously highlighted, numerous studies have previously referenced L-PRF using an array of protocols ranging from 2,500 to 3,000 RPM for 10 to 12 minutes on various-sized centrifuges, using various centrifugation tubes, at various rotor angulations, or using centrifuges fabricated from ranging manufacturing quality. In an attempt to increase the reproducibility of data, it is imperative that trade names such as L-PRF and A-PRF protocols be specifically applied solely when using their according centrifugation protocols (L-PRF = 2,700 RPM for 12 minutes on an IntraSpin Device; RCF-clot = 408 g, A-PRF+ = 1,300 RPM for 8 minutes on a Duo Quattro Centrifuge; RCF-max = 200 g). Attempts to reproduce an L-PRF membrane with a centrifugation device of different quality, using, different centrifugation tubes, on rotors of various dimensions and angulations should therefore be avoided in future studies. Recently, the concept of 'biological signature' has been proposed as a means to specifically address how one PRF membrane fabricated on one centrifugation device, may be entirely dissimilar from another if produced on a second centrifugation device with a different rotor size, rotor angulation, or produced in different tube sizes/compositions (even if RCF-clot/RCF-max values are comparable).[5] Owing to the quality and parameters differences in centrifugation devices (not all centrifugation systems are built with the same rotor sizes, tube angulations and quality manufacturing), it is imperative that should trademarks be used, their according centrifugation devices be appropriately used. This letter highlights the need to avoid inappropriately using trade names when specific proprietary protocols and devices are not explicitly used.

4 | CONCLUSIONS

When evaluating, and definitively when comparing medical devices and protocols, factual accuracy is of utmost importance. Within this letter, we highlight a minimum of six criteria that must be reported in all studies related to PRF to increase the transparency of research data in future publications. We further highlight the need to avoid inappropriately using trade names such as L-PRF and A-PRF when specific proprietary protocols and devices are not explicitly used. It is therefore important to highlight that the same protocol used in different clinics/patients increases the likelihood of having similar biological characteristics and clinical responses. We therefore encourage a stricter peer-review process regarding the report of RCF values in future scientific publications related to PRF to minimize future misrepresentations/inaccuracies in the field and increase the transparency of research related to platelet-rich fibrin. One should naturally only use devices and consumables that have been certified for use/application in patients as a class II medical device.

ACKNOWLEDGMENTS

All authors report no conflicts of interest related to this editorial.

REFERENCES

1. Castro AB, Meschi N, Temmerman A, et al. Regenerative potential of leucocyte- and platelet-rich fibrin. Part B: sinus floor elevation, alveolar ridge preservation and implant therapy. A systematic review. *J Clin Periodontol*. 2017;44:225-234.
2. Castro AB, Meschi N, Temmerman A, et al. Regenerative potential of leucocyte- and platelet-rich fibrin. Part A: intra-bony defects, furcation defects and periodontal plastic surgery. A systematic review and meta-analysis. *J Clin Periodontol*. 2017;44:67-82.
3. Ghanaati S, Herrera-Vizcaino C, Al-Maawi S, et al. Fifteen years of platelet rich fibrin (PRF) in dentistry and oromaxillofacial surgery: how high is the level of scientific evidence. *J Oral Implantol*. 2018;44:471-492.
4. Miron RJ, Zucchelli G, Pikos MA, et al. Use of platelet-rich fibrin in regenerative dentistry: a systematic review. *Clin Oral Investig*. 2017;21:1913-1927.
5. Dohan Ehrenfest DM, Pinto NR, Pereda A, et al. The impact of the centrifuge characteristics and centrifugation protocols on the cells, growth factors, and fibrin architecture of a leukocyte- and platelet-rich fibrin (L-PRF) clot and membrane. *Platelets*. 2018;29:171-184.
6. Kubesch A, Barbeck M, Al-Maawi S, et al. A low-speed centrifugation concept leads to cell accumulation and vascularization of solid platelet-rich fibrin: an experimental study in vivo. *Platelets*. 2018:1-12. https://doi.org/10.1080/09537104.2018.1445835
7. Wend S, Kubesch A, Orlowska 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;28:188.
8. Fujioka-Kobayashi M, Miron RJ, Hernandez M, Kandalam U, Zhang Y, Choukroun J. Optimized Platelet-Rich Fibrin With the Low-Speed Concept: growth Factor Release, Biocompatibility, and Cellular Response. *J Periodontol*. 2017;88:112-121.
9. Ghanaati S, Booms P, Orlowska A, et al. Advanced platelet-rich fibrin: a new concept for cell-based tissue engineering by means of inflammatory cells. *J Oral Implantol*. 2014;40:679-689.
10. Pinto N, Quirynen M. Letter to the editor: RE: Optimized platelet-rich fibrin with the low-speed concept: Growth factor release, biocompatibility, and cellular response. *J Periodontol*. 2019;90:119–121.

11. Miron R, Choukroun J, Ghanaati S. Controversies related to scientific report describing g-forces from studies on platelet-rich fibrin: necessity for standardization of relative centrifugal force values. *Int J Growth Factors Stem Cells Dent*. 2018;1:80-89.
12. Anwandter A, Bohmann S, Nally M, Castro AB, Quirynen M, Pinto N. Dimensional changes of the post extraction alveolar ridge, preserved with Leukocyte- and Platelet Rich Fibrin: a clinical pilot study. *J Dent*. 2016;52:23-29.
13. Pinto NR, Ubilla M, Zamora Y, Del Rio V, Dohan Ehrenfest DM, Quirynen M. Leucocyte- and platelet-rich fibrin (L-PRF) as a regenerative medicine strategy for the treatment of refractory leg ulcers: a prospective cohort study. *Platelets*. 2018;29:468-475.
14. Temmerman A, Vandessel J, Castro A, et al. The use of leucocyte and platelet-rich fibrin in socket management and ridge preservation: a split-mouth, randomized, controlled clinical trial. *J Clin Periodontol*. 2016;43:990-999.
15. Choukroun J, Diss A, Simonpieri A, 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-e60.
16. Dohan DM, Choukroun J, Diss A, et al. 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-e44.
17. El Bagdadi K, Kubesch A, Yu X, et al. Reduction of relative centrifugal forces increases growth factor release within solid platelet-rich-fibrin (PRF)-based matrices: a proof of concept of LSCC (low speed centrifugation concept). *Eur J Trauma Emerg Surg*. 2017. https://doi.org/10.1007/s00068-017-0785-7
18. 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;44:345-351.
19. Kobayashi E, Fluckiger L, Fujioka-Kobayashi M, et al. Comparative release of growth factors from PRP, PRF, and advanced-PRF. *Clin Oral Investig*. 2016;20:2353-2360.
20. Miron RJ, Fujioka-Kobayashi M, Hernandez M, et al. Injectable platelet rich fibrin (i-PRF): opportunities in regenerative dentistry. *Clin Oral Investig*. 2017;21:2619-2627.
21. Beck DJ, Bibby BG. A centrifugal technique of measuring food retention. *J Dent Res*. 1961;40:148-160.
22. Rickwood D. *Centrifugation: A Practical Approach*. Oxford, UK: IRL Press; 1984.

How to cite this article: Miron RJ, Pinto NR, Quirynen M, Ghanaati S. Standardization of relative centrifugal forces in studies related to platelet-rich fibrin. *J Periodontol*. 2019;90:817–820. https://doi.org/10.1002/JPER.18-0553