

# Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF

# Vilca-Molina Frank<sup>1</sup>, Cámara-Cabello Dora<sup>2</sup>, Paredes-Muñoz Gilmar<sup>1</sup>, Ponce-Soto Luis<sup>3</sup> and Obando-Pereda Gustavo<sup>3\*</sup>

<sup>1</sup>Dentistry School, Universidad Catolica de Santa Maria, Arequipa, Peru <sup>2</sup>Dentistry School, Universidad Cientifica del Sur, Lima, Peru <sup>3</sup>Protein Chemistry Laboratory, Universidad Catolica de Santa Maria, Arequipa, Peru

\*Corresponding Author: Obando-Pereda Gustavo, Protein Chemistry Laboratory, Universidad Catolica de Santa Maria, Arequipa, Peru.

Received: March 23, 2023; Published: April 08, 2023

# Abstract

**Aim:** The aim of this study was observe the influence of fasting and non-fasting in the weight, time of agglutination, and volume of L-PRF and I-PRF.

Materials and Methods: L-PRF and I-PRF were obtain from subjects in fasting and non-fasting.

**Results:** Fasting state showed better characteristics in volume and weight for L-PRF and I-PRF (p < 0.05) and the time to clot formation in I-PRF was longer in fasting state (p < 0.05). The data showed that volume, weight and time of coagulation for L-PRF and I-PRF is better in fasting state (P < 0.05).

**Conclusion:** It can be concluded that fasting state promote better characteristics for extraction of L-PRF and I- PRF than non-fasting state.

**Clinical Significance:** The present study reveals the importance of L-PRF and I-PRF extraction in fasting state for patients that requires oral surgeries because the great characteristics in volume, weight and time of clot coagulation.

Keywords: Fasting; Non-Fasting; Leukocyte Platelet Rich Fibrin; Injectable Platelet Rich Fibrin

# Introduction

Wound healing is the major goal in all regeneration therapies of either hard or soft tissues in dentistry and medicine, to provide fast and enhanced healing process to the patients [1-3]. The process of wound healing is a physiological response to tissue injuries that involves many types of cells, biological mediators and cells signals that are capable to produce the migrations of special cells to injured tissue [4,5] to enhance tissue regeneration. Many techniques has been propose to provide rapid tissue healings and regenerations using platelets from blood clot rich in cytokines and growth factors [3].

In 1998, Marx and collaborators found the positive effects of platelet rich plasma (PRP) on bone regeneration in combination with many techniques as autologous bone [6,7]. As this technique use a biological or chemical anticoagulant for its preparation, investigators has been propose another technique that do not use any anticoagulant, obtained a clean clot of fibrin without any biochemical blood

*Citation:* Obando-Pereda Gustavo., *et al.* "Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF". *EC Dental Science* 22.5 (2023): 16-21.

manipulation [8]. In 2001, Choukroun and co-workers introducing the protocol to obtain the leukocyte platelet rich fibrin (L-PRF) using freshly blood without any anticoagulant, been a completely autologous platelet concentrate [9]. This protocol consist in obtain freshly blood, drawn in a glass/glass based collection tubes follow by a centrifugation at 2700 - 3000 rpm for 12 minutes to obtain three layers: a red blood corpuscles at the bottom, PRF and platelet poor plasma at the top [1,9].

Macroscopically, PRF, is composed of yellow fibrin clot; microscopically consist in a 3D structure composed by a dense polymerized fibrin strands and at the junction of red zone, lymphocytes are present along with platelets clusters [3,8]. The biological activity of PRF to contribute to wound healing include platelet derived growth factors (PDGF), transforming growth factor-b (TGF-b), vascular endothelial growth factor (VEGF). Insulin like growth factor-1 (IGF-1), etc [8,10]. In this fact, PRF is entirely autologous in compare with PRP, and no contains any additives, been the PRF formation is completely physiological [8].

Several modification of PRF techniques have been suggest, related to the revolution in rpm or time as well in the material of tube collection [3]. This modifications are known as follows: Advance platelet rich fibrin (A-PRF) [11,12] and Injectable platelet rich fibrin (I-PRF) [13]. A-PRF is produce by a slower speed of centrifugation (1500 rpm) for almost 14 minutes as suggest Ghanaati [11] or 1300 rpm for 8 minutes as suggest Fujioka-Kobayahi [12]. This protocol aimed to increase the number of cells that contain PFR matrix and gain amounts of biological growth factors release [12]. This technique present some studies with contradictory results as poor polymerization [14] and growth factor release in compare with L-PRF [3].

Diet is a priority item for many effects, direct and indirect, of health including a haemostatic system [15]. Many studies suggest that the fatty dietary provoke an alteration in the coagulation and platelet function [15-18], however other studies present contradictory observations [15,19]. Indeed, dietary effects on platelet function and coagulation still not well characterized also their clinical management are unpredictable [15]. In other way, fibrin clot has presented changes in the 3D structure, been denser in fatty dietary and in compare with a light diet [15].

# Aim of the Study

The aim of this study is observe the influence of fasting and non-fasting in the weight, time of agglutination, and volume of L-PRF and I-PRF.

## **Materials and Methods**

# Study design

Blood samples of fifteen healthy patients were collected in fating and non-fasting state and L-PRF and I-PRF protocol extraction were performed. Clot volume and weight were measured and the time of clot formation of I-PRF were measured in fasting and non-fasting state.

#### **Subjects**

Fifteen healthy dentistry student males (22 - 24 years old) of Facultad de Odontología de la Universidad Católica de Santa María without history of medication consumption, with no evidence of systemic disease during the 60 days prior the study, not present obesity were included. All individuals signed the informed consent. The procedure is well approve by the Ethical Committee of Universidad Católica de Santa María N°211404.

# **Blood collection**

After 6 hours of last meal (fasting state), followed a skin disinfection, 20 mL of fresh blood were collected using a 20 mL syringe: ten mL of this blood were used to obtain L-PRF and the other 10 mL were used to obtain I-PRF. Immediately, the volunteers received an fatty diet equivalent to 1000 calories. One hour after the consumption of food, a non-fasting state, 20 mL of fresh blood were collected to obtain L-PRF and I-PRF and I-PRF.

*Citation:* Obando-Pereda Gustavo., et al. "Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF". EC Dental Science 22.5 (2023): 16-21.

17

#### L-PRF and I-PRF obtainment and processing

The obtainment of L-PRF was perform as described by Chorukroun; briefly, 10 mL of fresh blood were place in a non-coated glass tube, and quickly subjected to centrifugation at 2700 rpm for 12 minutes [20]. After the centrifugation, PRF was collected carefully and separate of the red phase [14,20]. The fibrin clot were place in glass-graduated cylinder (previously weighed) to verify the weight of the clot and volume obtained.

I-PRF was obtain as described by Mourao and Shah; briefly, 10 ml of fresh blood were placed in a non-coated glass tube and were subjected to centrifugation at 2700 rpm for 2 minutes [3,13]. After the centrifugation, the orange color area (I-PRF) were obtained using a 10 mL syringe without mixture the phases [13]. The I-PRF were place in a glass-graduated cylinder (previously weighed) to verify the time of I-PRF agglutination, weight and volume.

#### Statistical analysis

Data were expressed as mean ± SD. T-Student were used to compare the mean values to compare the fasting and non-fasting state to volume and weight for L-PRF, and volume, time and weight to I-PRF (GraphPad Prism 8V, CA, USA). Differences with P < 0.05 were consider statistically significant.

# Results

#### Influence of fasting and non-fasting state on L-PRF obtained

Volume in mL of L-PRF obtained from fasting state is great significant  $(3.745 \pm 0.53)$  to compare to the non-fasting state  $(3.309 \pm 0.41)$ p = 0.437 (Figure 1a). Weight in grams of L-PRF obtained from fasting state is great significant  $(3.264 \pm 0.54)$  to compare to the non-fasting state (2.808 ± 0.49) p = 0.474 (Figure 1b).



Figure 1: The L-PRF clot obtained present more volume (a) and weight (b) in fasting state than non-fasting state, p < 0.05.

# Influence of fasting and non-fasting state on I-PRF obtained

Volume in mL of L-PRF obtained from fasting state is great significant  $(1.609 \pm 0.43)$  to compare to the non-fasting state  $(1.067 \pm 0.29)$  p < 0.0018 (Figure 2a). Weight in grams of L-PRF obtained from fasting state is great significant  $(0.935 \pm 0.28)$  to compare to the non-fasting state  $(0.561 \pm 0.20)$  p < 0.0028 (Figure 2b). Time in seconds of I-PRF obtained from fasting state is great significant  $(5.923 \pm 1.25)$  to compare to the non-fasting state  $(3.615 \pm 0.86)$  p < 0.0001 (Figure 2b).

*Citation:* Obando-Pereda Gustavo., et al. "Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF". EC Dental Science 22.5 (2023): 16-21.

## Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF



Figure 2: The I-PRF sera obtained present more volume (a) and weight (b) and more time for fibrin clot formation (c) in fasting state than non-fasting state, p < 0.05.

# Discussion

Platelet concentrates treatment are fully used to many surgical and aesthetics procedures to accelerate wound healing mainly [8,9]. These procedures are perform in privates and publics clinics as simple interventions to improve the wound healing in time and quality [1]. L-PRF and I-PRF are techniques that used the secondary via of coagulation for the production of fibrin clot, a main component to release chemokines, cytokines and many growing factors to enhance the cell migration, differentiation and maintenance of cells responsible for regeneration and wound healing [1,13,21]. The obtainment of L-PRF and I-PRF needs a centrifugation that differ in G (rpm) and time [9,13,20]. However, exist a controversial issue in this procedure, if the fasting influences in the weight, volume and time of PRF extraction, important characteristics to improve to maximum the PRF clot.

Until now, in literature, no exist any explanation about the fasting influence in obtain of L-PRF and I- PRF. In the present study, it was demonstrated that fasting presents better conditions for volume and weight for both PRF techniques and the time is more extends for the clot obtaining in I-PRF. However, exist one report which indicate, briefly in methodology for the PRGF obtaining, that patients do not eat fatty food one day before the procedure and the patients have to be in fasting [21].

*Citation:* Obando-Pereda Gustavo., et al. "Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF". EC Dental Science 22.5 (2023): 16-21.

Many explanations exist in the influence of dietary and blood coagulation factors. Some studies explain a positive relationship in the increased of VIIc factor in a postprandial state [22-26] that can be observe a good fibrinogen concentration and a good modulation on fibrin clot structure [15]. However, this data do not explain the difference in volume and weight obtain of fibrin clot in fasting and non-fasting state showed in the present study. Other studies did not showed any relationship between dietary and coagulation [15,17,25,26]. In this manner, the dietary influence in the fibrin clot obtain for L-PRF and I-PRF is still unclear.

# Conclusion

Within limits of this study, it can be concluded that fasting state promotes better obtaining of volume, weight of L-PRF and I-PRF and time of formation of clot in I-PRF in compare with the non-fasting state.

# **Conflict of Interest**

The authors declare there are no conflict of interest exists.

# **Bibliography**

- 1. Karimi K., et al. "The Benefits of Platelet-Rich Fibrin". Facial Plastic Surgery Clinics of North America 27.3 (2019): 331-340.
- Kobayashi E., et al. "Comparative release of growth factors from PRP, PRF, and advanced- PRF". Clinical Oral Investigations 20.9 (2016): 2353-2360.
- 3. Shah R., *et al.* "An Update on the Protocols and Biologic Actions of Platelet Rich Fibrin in Dentistry". *The European Journal of Prosthodontics and Restorative Dentistry* 25.2 (2017): 64-72.
- 4. Miron RJ., *et al.* "Platelet-Rich Fibrin and Soft Tissue Wound Healing: A Systematic Review". *Tissue Engineering Part B, Reviews* 23.1 (2017): 83-99.
- 5. Qing C. "The molecular biology in wound healing and non-healing wound". *Chinese Journal of Traumatology = Zhonghua Chuang Shang Za Zhi* 20.4 (2017): 189-193.
- 6. Marx RE., *et al.* "Platelet-rich plasma: Growth factor enhancement for bone grafts". *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontics* 85.6 (1998): 638-646.
- 7. Oryan A., et al. "Platelet-rich plasma for bone healing and regeneration". Expert Opinion on Biological Therapy 16.2 (2016): 213-232.
- 8. Dohan DM., *et al.* "Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part II: platelet-related biologic features". *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontics* 101.3 (2006): e45-e50.
- 9. Dohan DM., *et al.* "Platelet-rich fibrin (PRF): a second-generation platelet concentrate. Part I: technological concepts and evolution". *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontics* 101.3 (2006): e37-e44.
- 10. Kim TH., *et al.* "Comparison of platelet-rich plasma (PRP), platelet-rich fibrin (PRF), and concentrated growth factor (CGF) in rabbitskull defect healing". *Archives of Oral Biology* 59.5 (2014): 550-558.
- 11. Ghanaati S., *et al.* "Advanced platelet-rich fibrin: a new concept for cell-based tissue engineering by means of inflammatory cells". *The Journal of Oral Implantology* 40.6 (2014): 679-689.
- 12. Fujioka-Kobayashi M., *et al.* "Optimized Platelet-Rich Fibrin with the Low-Speed Concept: Growth Factor Release, Biocompatibility, and Cellular Response". *Journal of Periodontology* 88.1 (2017): 112-121.

*Citation:* Obando-Pereda Gustavo., *et al.* "Influence of Fasting and Non-Fasting on Extraction of L-PRF and I-PRF". *EC Dental Science* 22.5 (2023): 16-21.

20

- 13. Mourao CF., *et al.* "Obtention of injectable platelets rich-fibrin (i-PRF) and its polymerization with bone graft: technical note". *Revista do Colegio Brasileiro de Cirurgioes* 42.6 (2015): 421-423.
- 14. Dohan Ehrenfest DM., *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* 29.2 (2018): 171-184.
- 15. Pieters M., et al. "Diet and haemostasis a comprehensive overview". Blood Reviews 29.4 (2015): 231-241.
- 16. Al-Attas OS., *et al.* "Association of dietary fatty acids intake with pro-coagulation and inflammation in Saudi Adults". *Asia Pacific Journal of Clinical Nutrition* 23.1 (2014): 55-64.
- 17. Miller GJ. "Dietary fatty acids and the haemostatic system". Atherosclerosis 179.2 (2005): 213-227.
- 18. Olas B. "Dietary Supplements with Antiplatelet Activity: A Solution for Everyone?". Advances in Nutrition 9.1 (2018): 51-57.
- 19. Yang ZH., *et al.* "Dietary supplementation with long-chain monounsaturated fatty acid isomers decreases atherosclerosis and alters lipoprotein proteomes in LDLr(-/-) mice". *Atherosclerosis* 262 (2017): 31-38.
- 20. Choukroun JAF., et al. "Une opportunité en paro-implantologie: Le PRF". Implantodontie 42 (2001): 55-62.
- 21. Anitua E. "The use of plasma-rich growth factors (PRGF) in oral surgery". *Practical Procedures and Aesthetic Dentistry: PPAD* 13.6 (2001): 487-493.
- 22. Duttaroy AK. "Postprandial activation of hemostatic factors: role of dietary fatty acids". *Prostaglandins, Leukotrienes, and Essential Fatty Acids* 72.6 (2005): 381-391.
- 23. Silveira A. "Postprandial triglycerides and blood coagulation". *Experimental and Clinical Endocrinology and Diabetes: Official Journal, German Society of Endocrinology [and] German Diabetes Association* 109.4 (2001): S527-532.
- 24. Van Der Toorn FA., *et al.* "Glucose metabolism affects coagulation factors: The NEO study". *Journal of Thrombosis and Haemostasis: JTH* 17.11 (2019): 1886-1897.
- 25. Mutanen M., et al. "Fats, lipids and blood coagulation". Current Opinion in Lipidology 12.1 (2001): 25-29.
- 26. Miller GJ. "Effects of diet composition on coagulation pathways". The American Journal of Clinical Nutrition 67.3 (1998): 542S-545S.

Volume 22 Issue 5 May 2023 © All rights reserved by Obando-Pereda Gustavo., *et al.*  21