

# Autologus Barriers, Fillers and Growing Factors: Using the Secondary Pathway of Haemostasis in Dental Clinics-Report of Four Clinical Cases

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## Abstract

Haemostasis is an important system that maintain the hydrolytic balance of the body. The secondary pathway of this system produce a fibrin network to improve coagulation and regeneration. Platelet rich in fibrin, use the secondary pathway of haemostasis to isolate large quantities of fibrin in vitro, for clinical uses. In dental clinic, for improve bone regeneration, the PRF protocol it has become the principal toll for these purposes, leading a predictable bone regeneration and rapid osseointegration.

**Keywords:** Residual root extraction; Wisdom molar extraction; Osteointegration; Platelet plug

## Introduction

Haemostasis is a unique and important system of physiology to maintain the hydrolytic balance in the body, stop bleeding [1]. When an injury occurs, a vasoconstriction lead the beginning of the primary haemostasis reducing the amount of blood flow through the area and limits the amount of blood loss where the platelets are activated to form a platelet plug [1,2]. To address the tissue regeneration a secondary path of haemostasis must be activated, where a blood clotting must be done [3]. This blood clotting needs fibrin threads that act as a glue for the sticky platelets [4,5]. As the fibrin mesh begins to form, the blood is also transformed from a liquid to a gel like substance through involvement of clotting and growing factors and pro-coagulants [1,3,5]. The coagulation process is useful in closing up and

maintaining the platelet plug on larger wounds to tissue regeneration [6], in this fact, formation of a stable clot that is capable of withstanding the pressure in a blood vessel is essential to prevent bleeding and to promote wound healing [6,7]. Therapies using fibrin has been proposed to increase the tissue regeneration, and has been used for many conditions, like oral tissue regeneration [8-11], and plastic surgeries mainly [12,13]. Platelet rich in fibrin belongs to a new generation of platelet concentrates characterized by a simple preparation and, the most important item, the conservation of all biological components of blood [14]. Choukroun first developed the PRF in France for use in oral surgery [14]. This technique avoids any chemical anticoagulant or coagulant in the preparation, simplifying the technique, letting the natural polymerization of blood, producing a 3-dimensional organization of a fibrin

network [15].

The PRF process leads to engage the accumulation of many biological healing factors like cytokines, glycanic chains and structural glycoproteins (fibronectin) and immunity promoters trapped in the colloidal suspension between the fibrin network during centrifugation; these biological factor are available *in situ* for a convenient period, to lead the cells start the cicatricial matrix remodeling having well known synergetic effects on healing processes [16,17]. Platelet derived growth factors (PDGFs) are the first growing factors to be present in a wound initiating the connective tissue healing, bone regeneration and repair [18]. This factor is crucial in the migration, proliferation and survival of mesenchymatous cells linages, where perform well mitogenesis, angiogenesis and macrophage activation [19,20]. Moreover, PRF, contain IL-4, a healing cytokine, who during inflammatory phenomena supports healing process by modulation of inflammation and immune control [21]. Other important biological factor is the vascular endothelial growth factor (VEGF) considered as a master regulatory molecule for angiogenesis process, and plays a direct role in the control of endothelial cell behaviors, such proliferation, migration, specialization and survival enhancing the wound healing [17,21].

In this context, what happen if a large amount of fibrin clot is placed in a surgical wound?

## Clinical Cases

### Clinical case 1: Residual root extraction

A 21 years old female was come to the Periodontics and Dental Implantology course, for evaluation by a residual root referent to right maxillary second premolar.

**Presurgical Therapy:** The surgical procedure was explained to the patient and the informed consent was obtained. Pharmacology preparation of the patient included ketorolac (90mg), dexamethasone (4mg) and antibiotic prophylaxis with amoxicillin (2g) one hour before the procedure.

**PRF preparation:** The protocol was performed as described by Dohan, et al. [15]. Briefly, 10 mL of blood was drawn in 15 mL test tube without an anticoagulant and centrifuged immediately using a tabletop centrifuge (GREETMED CENTRIFUGE mod. GT119-100T, China) at 3000 rpm for 10 minutes.

The resultant product consists of the following three

layers:

- Top most layers consist of cellular plasma.
- PRF clot in the middle.
- Red blood corpuscles at the bottom.

With the absence of an anticoagulant, blood begins to coagulate as soon as is exposed to glass surface of test tube. Therefore, for successful preparation of PRF, blood must be collected and immediately centrifugation leads the clotting cascade initiated. PRF was obtain in the form of gelatinous plasma. Surgical procedure: after proper isolation of the surgical field, the operative site was anaesthetized using 72 mg of lidocaine with epinephrine (1:80000). An on line incision was made along the alveolar ridge soft tissue to retail a mucoperiostial flap. The residual root was extracted and the alveolar socket was curettage to remove any granulose and infected tissue. Immediately to curettage, the PRF was inserted in the alveolar socket. An autologous barrier was produced from PRF and this was putted on alveolar bone closed the alveolar socket. Soft tissue was approached and sutured to enhance a first intention wound healing. A periapical x-ray was performed immediately ending the surgical procedure and at 30 days (Figure 1).

**Postoperative healing:** There was not post-operative complication and healing was satisfactory. Radiographic control suggested the formation of an osseous matrix in all alveolar socket at 30 days.

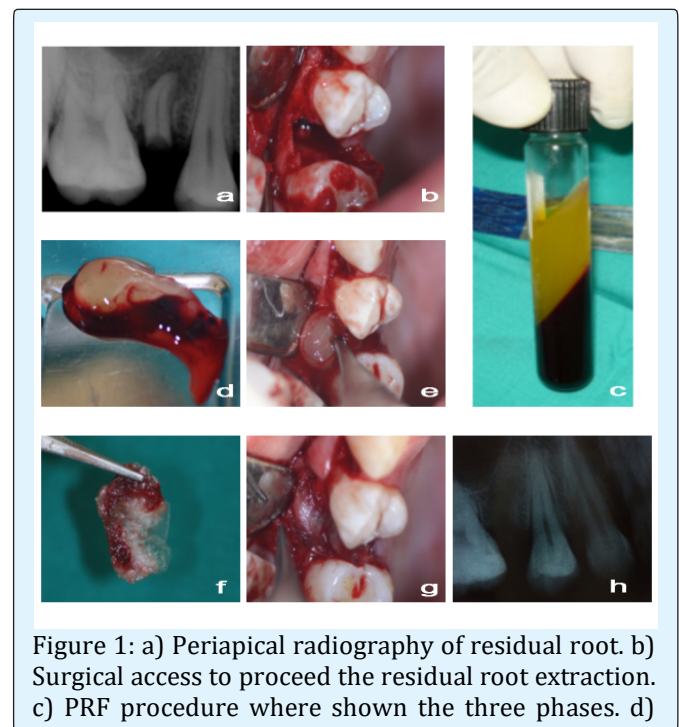


Figure 1: a) Periapical radiography of residual root. b) Surgical access to proceed the residual root extraction. c) PRF procedure where shown the three phases. d)

PRF clot were isolated and it cut in two parts. e) One part it was placed into the alveolar socket. f) The other part was performed an autologous barrier. g) The autologous barrier was placed to close the alveolar socket with PRF. h) Periapical radiography thirty days after surgery showed an alveolar socket fill with bone matrix.

### Clinical case 2: Wisdom molar extraction

A 27 years old female was come to the Periodontics and Dental Implantology course, for evaluation by pain referent to left mandibular third molar.

**Presurgical therapy:** The surgical procedure was explained to the patient and the informed consent obtained. Pharmacology preparation of the patient included ketorolac (90mg), dexamethasone (4mg) and antibiotic prophylaxis with amoxicillin (2g) one hour before the procedure.

**Surgical procedure:** After proper isolation of the surgical field, the operative site was anaesthetized using 72 mg of lidocaine with epinephrine (1:80000). An Avellanal incision was performing to expose the coronal part and alveolar bone following by an osteotomy to extract the third molar. The alveolar socket was curettage to remove any granular tissue. Following the extraction, a PRF clot was placed into the alveolar socket and soft tissue was approached and sutured to enhance a first intention wound healing.

**Postoperative healing:** There was not post-operative complication and healing was satisfactory. Radiographic control suggested the formation of an osseous matrix in all alveolar socket at 30 days (Figure 2).

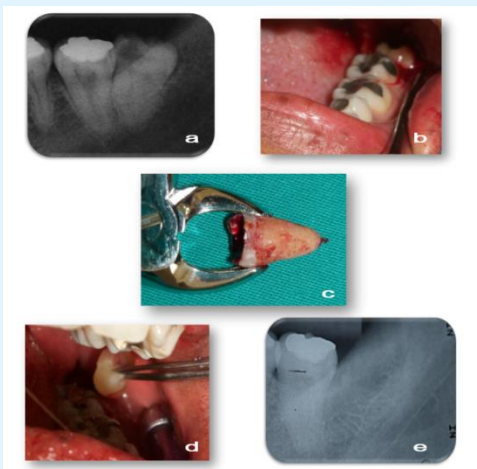


Figure 2: a) Periapical radiography of wisdom tooth.

b) Surgical access to proceed the wisdom tooth extraction. c) Wisdom tooth with fused roots. d) PRF was placed into the alveolar socket. e) Periapical radiography thirty days after surgery showed an alveolar socket fill with bone matrix.

### Clinical case 3: Rapid osteointegration in dental implants

A 18 years old male was come to the Private Practice, for evaluation by periodontosis of right superior central incisor. Because the vestibular bone cortical presented a fissure and the presence of parafunctional habits two stages implant rehabilitation was elected as treatment.

**Presurgical therapy:** The surgical procedure was explained to the patient and the informed consent obtained. Pharmacology preparation of the patient included ketorolac (90mg), dexamethasone (4mg) and antibiotic prophylaxis with clindamycin (600 mg) one hour before the procedure (Figure 3).

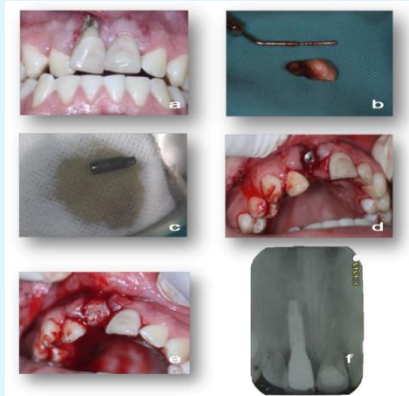


Figure 3: a) Central incisor lesion with apical decay and receding gum. b) Surgical access to proceed the central incisor extraction. c) PRF procedure where performed and dental implant was treated with PRF supernatant. d) Bone drilling was performing and was placed the treated dental implant. e) PRF autologous barrier was done and was placed into the alveolar socket protecting the dental implant. f) Sixty-days after surgery was performed the rehabilitation with ceramic crown and periapical radiography was taken suggesting the osseointegration.

**Surgical procedure:** After proper isolation of the surgical field, the operative site was anaesthetized using 72 mg of lidocaine with epinephrine (1:80000). A clean extraction was performing using a periosteal to conserve alveolar bone diameter. Sequence drill was done to reach a 3.5 mm diameter, to place a conic 4.0 x 11 mm internal hexagon

dental implant (Conexao, Sao Paulo, Brazil). Before dental implant place a PRF was performing and the fibrin clot was extracted. Dental implant was placed into de fibrin clot to capture any rich biochemical and biological factors in the surface. After that, the implant was immediately placed into the bone.

**Postoperative healing:** There was not post-operative complication and healing was satisfactory, radiographic and antitorque control suggested the osseointegration at two months.

**Prosthetic rehabilitation:** Prosthetic rehabilitation was performed using a porcelain crown.

#### Clinical case 4: Filling large bone defect with implant placement “Obando’s file technique”

A 48 years old female was come to the Private Practice for large coronal destruction of first left mandibular molar.

**Presurgical therapy:** The surgical procedure was explained to the patient and the informed consent obtained. Pharmacology preparation of the patient included ketorolac (90mg), dexamethasone (4mg) and antibiotic prophylaxis with amoxicillin (2g) one hour before the procedure.

**Surgical procedure:** after proper isolation of the surgical field, the operative site was anaesthetized using 72 mg of lidocaine with epinephrine (1:80000). An incision was performing and the molar was extracted. The mesial and distal roots were fusionated resulting a large alveolar socket. The inferior nerve canal was a 2mm to the apical portion of alveolar socket. The alveolar socket was curettage to remove any granular tissue. A 60 mL amount of blood was extracted from the patient and the PRF was performed. Following the extraction, a PRF clot was placed around of Bicon implant and the implant was placed in to the alveolar socket with primary stability less than 20 N. Another PRF clot was placed above of dental implant to close the alveolar socket.

**Postoperative healing:** there was not post-operative complication and healing was satisfactory. Radiographic control suggested the formation of an osseous matrix in all alveolar socket and anosseointegratoin of implant was observed at fourth month.



Figure 4: a) Periapical radiography of first inferior molar showed large coronal destruction with dental pulp compromise. b) Surgical access to proceed the molar extraction, it was present a fused roots. c) PRF procedure where performed and fibrin clot was placed around of dental implant. d) Dental implant with fibrin clot were placed into the alveolar socket reaching a primary stability less than 20N, fibrin clot and autologous barrier was done and was placed into the alveolar socket protecting the dental implant. e) Periapical radiography was taken fourth months after surgery showed a bone healing suggesting the osseointegration. f) Immediately was performed the rehabilitation with ceramic crown.

#### Discussion

The PRF protocol is frequently used with very favourable results for growth of hard and soft in the specialist dental clinic tissues. In implantology, it is also frequently used because of their biological factors that accelerate the processes of osseointegration [22] and especially accelerate bone formation in extraction sockets for future implant placement [23]. The case N<sup>o</sup>1 and N<sup>o</sup>2 PRF show the benefits of rapid bone regeneration in bone sockets after teeth extraction. This observation is in accordance with Anwandter (2016) [24] who describe well ridge preservation after teeth extraction [24]. This event can be explained by the realising of growth factors mainly and cytokines immersed in the fibrin clot that can control the inflammatory response and the regenerative properties of immune system to modulate of cellular migration and proliferation, accelerating the bone healing [25,26]. In the case of third molar, PRF causes a periodontal health distal to second molar avoiding posterior inflammation [27].



The case N<sup>o</sup> 3 not rich plasma was placed into fibrin in the socket, but the biological components clot implant transferred, leaving this in direct contact for the absorption of such biological factors may consequently form bone quickly by the expression BMPs [28]. Thus and subjectively, it can explain the accelerated osseointegration obtained in two months by activation of BMPs by the presence of the extracted biological factors PRF [28,29], these results are in agreement with those observed by Oncu et al. [22]. In case No. 4, fill a large alveolus was perform with concomitant implant placement without sufficient primary stability for osseointegration. According to some reports literature [22], the PRF is used to increase the primary stability of the implant filling the bone bed first with PRF and then place the implant. In this case, the clot PRF was prepared around the implant and placed in the socket with a primary stability of 30N (only was given a direction to the implant with respect to the future prosthesis) and then completely fills the rest of the alveolus with PRF. In our teaching practice as a private, we perform the Obando technique to increase primary stability and filler large bone defects.

All cases showed the rapid bone healing and bone regeneration of PRF protocol in dental surgery and dental implant. Preserving bone quantity and quality for future implant rehabilitation are the most concern topic in implant surgery planning [10,30-32]. Specialist employed commercial bone to fill bone socket after dental extraction with the consideration of healing time of this socket between 4 or 6 months [33-35]. However, employing fibrin clot, the bone quality and quantity are same or more than commercial substitutes in less of time [24,36]. The rapid osseointegration is directly related with the dental implant surface treatment and the biological growing factor as BMPs [37,38]. Some studies showed a rapid osseointegration of dental implant when BMPs are using on implant surface before placed into the bone [37-39]. However, commercial BMPs are very expensive and the cares of these proteins are many to avoid its denaturation. PRF offers similarities results because presents many growing factors including pre-BMPs which promotes acceleration of bone healing in dental implant treatments shortening the osseointegration time [28,40]. In this fact, large quantity of PRF cause positives effects in wound healing leading predicable bone regeneration and rapid osseointegration in dental implant clinics.

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