

Tissue Engineering, Platelets Concentrates and its Role in Dental Implant Treatment

Ayoub AH^{1,2*}, Ramadan OR³ and Agbor MA⁴

¹President of the Egyptian Society of Oral Implantology, Egypt

²President of International Group for Oral Rehabilitation, Alexandria, Egypt

³Department of Oral pathology, Faculty of Dentistry, Alexandria University, Alexandria, Egypt

⁴Department of Dentistry, Universite des Montagnes, Bangangte, Cameroon, Egypt

***Corresponding Author:** Ayoub AH, President of the Egyptian Society of Oral Implantology, President of International Group for Oral Rehabilitation, Alexandria, Egypt.

Received: August 13, 2016; **Published:** September 06, 2016

Abstract

Introduction: Guided tissue regeneration allow for osseous regeneration prior to soft tissue migration into an area of interest. This is accomplished with the use of membranes that prevent the migration of the soft tissue element into the bony defects. The use of platelet concentrates is recent and its efficiency remains controversial. Several techniques for platelet concentrates are available however, their applications have been confusing because each method leads to a different product with different biology and potential uses. Pure platelet-rich plasma (PRP) and platelets rich fibrin (PRF) are the common types of platelets rich plasma used in Dentistry. The latest innovation of platelets concentrates (i.e. PRP and PRF) is the mineralized plasmatic matrix (MPM) also called sticky bone.

Methods: This is a descriptive clinical study carried out on 2 extraction sites of an adult male who presented in a private practice in Alexandria- Egypt. Platelet concentrates were prepared individually in the clinic by centrifuging collected blood samples to obtain PRF and MPM. These samples were placed in extraction sites immediately after dental extraction and bone samples collected from the sites for histological evaluation after 4 months.

Results: The first socket treated with MPM histologically showed decalcified sections with a mass of cancellous bone and areas of diverse degrees of maturation and numerous large lacunae containing osteocytes as a prominent feature. The bony trabeculae were lined by numerous osteoblasts with the bony interface showing areas of active deposition of woven bone. The second socket treated with PRF presented with a calcified section showing a mass of mature cancellous bone with prominent resting lines. The bony trabeculae demonstrated multiple osteocytes in large lacunae. Few viable osteoblasts were seen lining the bony trabecula.

Conclusion: The use of bone filler combined with fibrin, platelets and leukocytes in the form of MPM have shown a better histologic evidence of hard bone formation as evidenced by its high osteoblastic activity and maturation than the use of PRF as sole filling material for the extraction socket after 4 months.

Keywords: Extraction; PRF; MPM; Socket preservation

Introduction

Guided tissue regeneration (GTR) is a technique currently used in dentistry for periodontal surgery, oral surgery, implant dentistry and reconstruction of maxillomandibular defects. The basic premise for this technique is to allow for osseous regeneration prior to soft tissue migration into the area of interest. This is accomplished with the use of membranes that prevent the migration of the soft tissue element into the bony defect [1]. Successfully osteo-integrated implants essentially require adequate quality and quantity of bone at the implant site.

Ridge defects develop as a result of surgery, trauma, infection, or congenital malformations. The goals of osseous replacement are maintenance of contour, elimination of dead space, and reduce postoperative infection; and thus enhance bony and soft tissue healing. The insufficient quantity of bone is due to tooth loss which results in rapid resorption of alveolar bone due to lack of intraosseous stimulation by periodontal ligament (PDL) fibers, for example, pneumatization of maxillary sinus following tooth loss [2].

Once a tooth is extracted, the alveolar ridge inevitably undergoes remodeling with associated resorption which diminishes the size of the ridge. It leads to compromises in the functional and aesthetic outcomes of implant and thus hard and soft tissue augmentations are needed [3,4].

Loss of alveolar bone may be attributed to factors such as endodontic pathology, periodontitis, facial trauma and aggressive maneuvers during extractions etc. Millions of teeth are still extracted annually which eventually end up in dental implant or prostheses replacements [5].

Bone grafting is a surgical procedure that replaces missing bone with material from patient's own body, an artificial, synthetic, or natural substitute. Bone grafting is possible because bone tissue has the ability to regenerate completely if provided the space into which it has to grow. As natural bone grows, it generally replaces the graft material completely, resulting in a fully integrated region of new bone [2].

Most extractions are done with no regard for maintaining the alveolar ridge [6,7]. Whether due to caries, trauma or advanced periodontal disease, tooth extraction and subsequent healing of the socket commonly result in osseous deformities of the alveolar ridge, including reduced height and reduced width of the residual ridge [7]. This is because after extraction, alveolar socket undergoes subsequent remodeling process, which is a means of natural additional atrophy. It begins as soon as the tooth is extracted and within a period of 3 months almost 50% alveolar ridge gets resorbed [8]. Additional loss of bone occurs when the extraction is not performed a traumatically.

The severity of the healing pattern may pose a problem for the clinician in 2 ways: it creates an esthetic problem in the fabrication of an implant-supported restoration or a conventional prosthesis and it may make the placement of an implant challenging [9]. However, it is possible to minimize such problems by simply carrying out ridge preservation procedures in extraction sockets using grafting materials with or without barrier membranes [10]. The various regenerative biomaterials used for socket augmentation are grafts, membranes, biologic modifiers, and platelet concentrates [5].

Platelet-rich fibrin (PRF) and the mineralized plasmatic matrix (MPM) are examples of platelet concentrates used for tissue regeneration in dentistry.

Various platelet-derived products or platelet concentrates have been introduced that act as biological mediators aiding the healing response. Platelet-rich fibrin (PRF) is one such product that has proved its worth and has edged past the others. The Choukroun's platelet rich fibrin has opened the flood-gates in the field of dentistry, majorly focusing on the improved healing and regeneration [11]. Thus PRF has also been tagged as a healing biomaterial [11].

Platelet-rich fibrin (PRF) belongs to a new generation of platelet concentrates with simplified processing without biochemical blood handling. It is a strictly autologous fibrin matrix containing a large quantity of platelet and leukocyte cytokines [12]. The use of platelet gel to improve soft and hard tissue regeneration is still a recent technique in dental Implantology and periodontology [13,14].

The mineralized plasmatic matrix (MPM) is a modification of the PRP and the PRF initially presented by Perisse and modified by El Moheb [15,16]. The advantage of the MPM is the integration of bone grafts particles inside the fibrin network that is not present in old autologous growth factors membranes in PRF or PRM. In fact, the bone grafting materials are prepared and mixed with the autologous growth factors to produce the MPM. This offers the MPM the positional stability [15,16] by stabilizing the bone particles, preserving its shape with subsequent "in situ" immobilization of the component of ridge preservation materials.

This article focuses on comparing the healing patterns of extraction sockets after filling them with PRF and MPM materials using histological laboratory analysis. Histological evidence was also reviewed to provide an in-depth understanding of the effect of 2 materials on accelerating the bone healing.

Methodology

This is a descriptive prospective clinical case study carried out on 2 extraction sites of an adult male who presented in a private practice in Alexandria-Egypt.

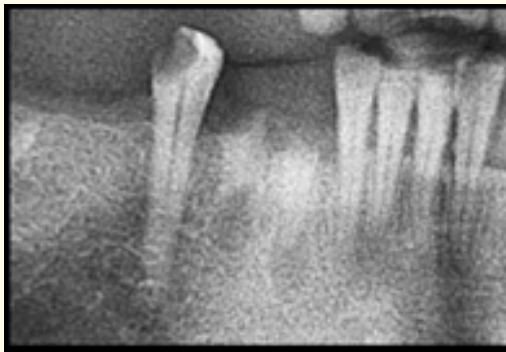
The study was carried out on a healthy 46 years' man who presented with fractured lower right lateral incisor (#41) and canine (#43) both teeth fractured up to the cervical margins. Recent dental history of the patient included placement of several implants. The patient also had a positive history of periodontal diseases and was a smoker.



On examination, the oral hygiene was poor, calculus was +++ (covering all the cervical surfaces of the posterior teeth), a good amount of buccal bone was present and a thin gingival biotype presented around the teeth. Periapical radiograph revealed vertical bone resorption.

Surgical Procedures and grafts preparations

Atraumatic extraction of the remaining part of the tooth was carried out without damaging the surrounding bone. Extractions were performed using the peristome to preserve the socket size and surrounding structures (Figure 2). Socket debridement and curettage was performed to eliminate remnants of periodontal fibers and necrotic tissues using a sharp curette.



Approximately 28ml of blood was collected from the patient's arm with a syringe and transferred to four 7ml centrifugal tubes without anticoagulant in (2 PRF tubes and 2 plain tubes). The blood samples were immediately centrifuged at 3000 rpm (approximately 400g according to Choukroun's calculations) for 10 minutes [5].

MPM Preparation

After 10 minutes of centrifugation, the 2 plain tubes presented 2 layers. The 1st layer (reddish in colour) at the bottom of the tube contained red blood cells (RBC's) and the second upper portion of the tube contained an amount of clear yellow plasma rich in leukocytes, platelets, mesenchymal stem cells and fibrinogen. The separation of the 2 layers is due to the differences in density of the blood components.

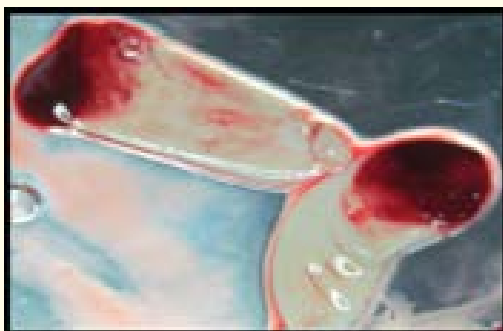
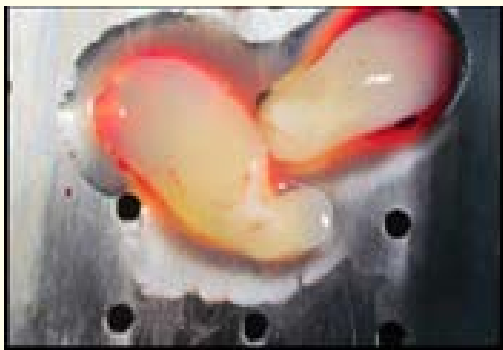
The 2nd layer was then mixed with the bone grafting material (A mix of 30% HA and 70% Tri calcium phosphate small granules manufactured by POLYSTOM) and a drop of patient blood from the extraction socket were mixed together in a sterile bowl. The drop of blood was added to provide the thrombin which will initiate the conversion of insoluble fibrinogen into soluble fibrin. After a couple of minutes, a homogenous mixture of fibrin network with integrated bone graft particles inside and the mixture is rich of platelets, leukocytes and mesenchymal cells. The MPM, which has been obtained, is placed in the extraction socket of the lateral incisor (#41). The edges of the mucosal flaps were approximated to each other and sutured using 3-0 Monocrylsutures.

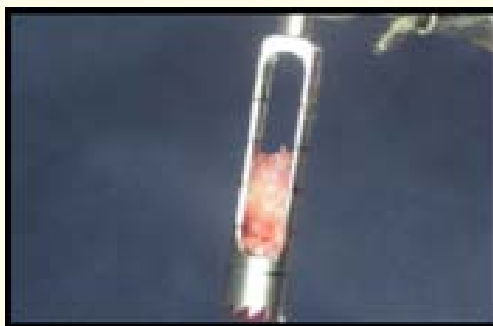
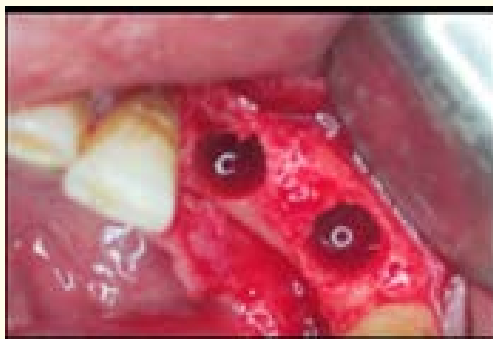
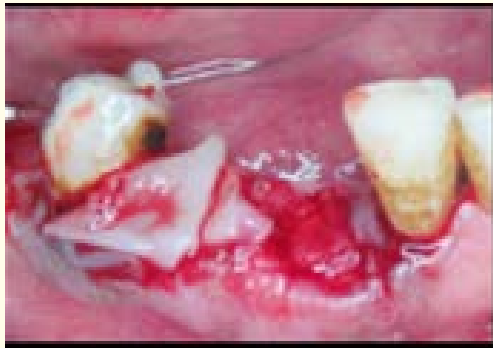


PRF Preparation

Within a few minutes, the absence of anticoagulant allowed the activation of the majority of platelets contained in the sample to trigger a coagulation cascade.

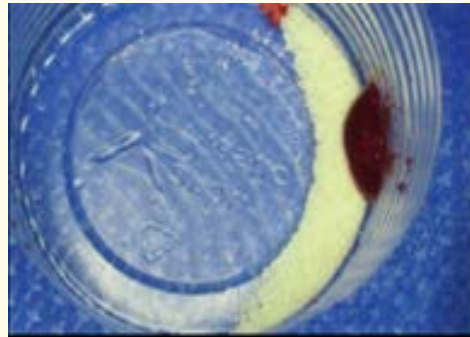
In the PRF tubes, fibrinogen is at first concentrated in the upper part of the tube until the effect of the circulating thrombin transforms it into a fibrin network [3]. A fibrin clot is then obtained in the middle of the tube just in the interface between the red corpuscles at the bottom and acellular plasma at the top.





Platelets are theoretically trapped massively in the fibrin mesh. The clot is removed from the tube and the attached red blood cells scraped off and discarded. The PRF clot is then placed on the grid in the PRF Box and covered with the compressor and lid. This produces an inexpensive autologous fibrin membrane in approximately one minute. The PRF Box is devised to produce membranes of constant thickness that remain hydrated for several hours and to recover the serum exudate expressed from the fibrin clots which is rich in the proteins vitronectin and fibronectin. The exudate collected at the bottom of the box may be used to store autologous grafts [4].

After removing the cover of PRF membranes obtained from 2 PRF clots (Figure 13), a specific tweezer was used to insert the PRF in the canine socket. Edges of the mucosal flaps were approximated to each other and sutured using 3-0 Monocrylsutures.



Healing was uneventful in both sites and the patient was followed up for 4 months postoperatively. During the follow-up, oral hygiene was reinforced and rinsing was carried out with 0.12% chlorhexidine.

Histological analysis

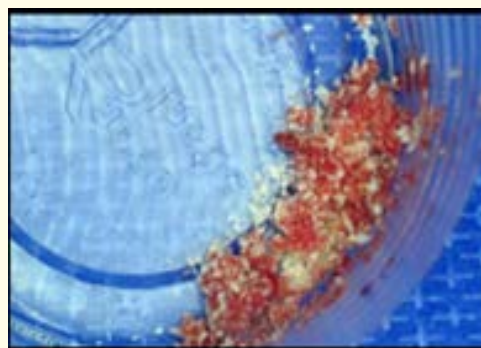
Four months later a small flap was raised and bone was harvested from the 2 sockets using a 3.2mm trephine drill and 2 implants micro dent GN3512 were inserted. The harvested bone was sent to the histopathology laboratory for histological evaluation.

Results

The results of this study was basically histological.

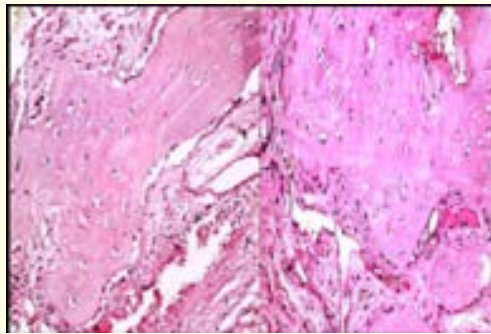
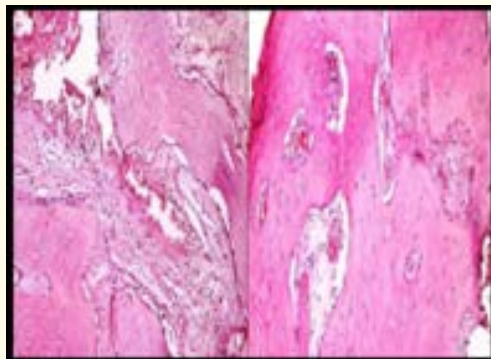
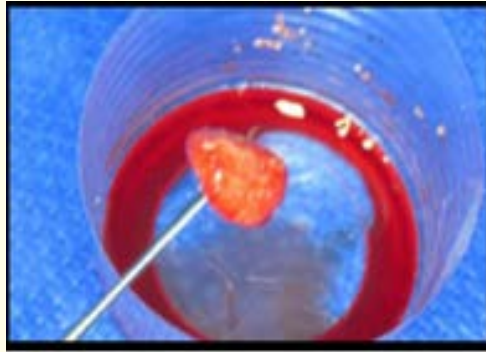
MPM sample analysis

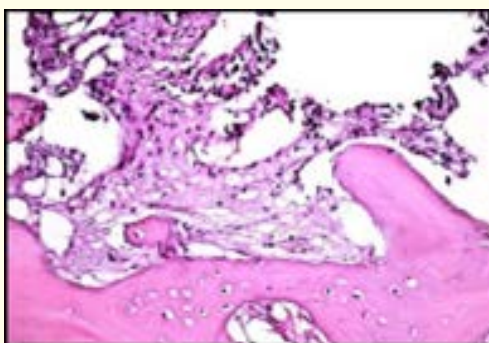
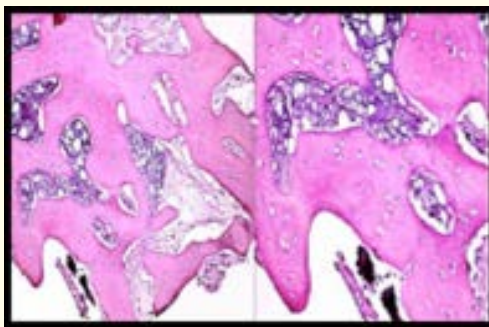
The first socket treated with MPM histological showed decalcified sections, and a mass of cancellous bone with areas of diverse degrees of maturation. It also presented with numerous large lacunae containing osteocytes as a prominent feature. The bony trabeculae were lined by numerous osteoblasts with the bony interface showing areas of active deposition of woven bone. Resting lines were seen in some areas and the bone marrow contained multiple capillaries and lymphatic vessels with mild inflammatory infiltrate.



PRF sample analysis histological study

Decalcified section showed a mass of mature cancellous bone with prominent resting lines. The bony trabeculae demonstrate multiple osteocytes in large lacunae. Few viable osteoblasts were seen lining the bony trabeculae. The bone marrow was clearly fibrotic with little vascularity and almost no chronic inflammatory cells infiltrating (Figure 15).





Discussion

Wound healing is a staged process which involves the activity of leukocytes and platelets. For this process to work efficiently, the platelets play a vital role. The growth factors present in platelets are important to guide the regenerating cells to the area of healing. Platelet-rich-fibrin (PRF) is one such material that holds on to these growth factors enmeshed in the fibrin network resulting in their sustained release over a period of time that can accelerate the wound healing process. With this knowledge, research has been carried out for a past few years for the clinical application of PRF. Various platelet concentrates have been studied including the platelet-rich-plasma (PRP). However, the short duration of cytokine release and its poor mechanical properties have resulted in the search of a new material with adequate properties for clinical application and ease of preparation. PRF has found a place in the regenerative field owing to its advantages over PRP [11].

Socket preservation is preserving the socket/ridge volume within the socket confines existing at the time of extraction [3]. Socket augmentation is increasing the socket/ridge volume beyond the socket confines or skeletal envelope existing at the time of extraction [17].

Because tooth extractions cause alveolar ridge resorption i.e. horizontal (buccal and lingual) and vertical, more resorption on buccal/facial side than on the lingual/Palatal and three-dimensional resorption leads to narrow ridge, reduced vertical height, and linguopalatal shifting of the long axis of the alveolar ridge [3]. These loss of bone do not only affect the quality of work of the clinician but also the esthetics of the patients as the stability and presentation of prosthesis in the patient's mouth can be compromised more especially the facial profile of the patients.

In contemporary dentistry, the quality and quantity of the bone is very important in defective ridge leads to incorrect placement of dental appliances like endosseous implants and inappropriate prosthetic fabrication with poor aesthetic outcome [3].

In the current study, the MPM specimen presented with a mass of cancellous bone with areas of diverse degree of maturation, this is because osteoblastic activity of the specimen is more prominent and active resulting in the deposition of woven bone while the PRF specimen presented with less osteoblastic activity resulting in fibrotic bone marrow with little vascularity.

In this study the plasma obtained after a single spin was rich in platelets, fibrinogen and monocytes. The fibrinogen is necessary for the formation of the MPM. It is this fibrinogen that will be transformed into fibrin network by thrombin obtained from blood extracted from the patient's wound [15]. Platelets have been reported to provide growth factors and the monocytes once activated by interleukin and this can enhance the production of BMP-2 [15]. BMP-2 is a bone morphogenetic protein that induces the bone formation. It is a high inductive protein [16].

The fibrin network that is produced in the MPM has several roles:

1. It serves as a mesh that will link all bone particles together to form a complex of made up of bone, platelets and monocytes.
2. It also serves as the scaffold needed by the bone to regenerate and also it is the pathway need by the cells to migrate to heal or to repair the wound.
3. This fibrin network or the extracellular matrix allows the elasticity of the MPM, permit the shaping and the remodeling of the MPM and facilitate the cells migrating inside the MPM between the particles. These could explain why MPM has the potential of forming a harder and matured bone within 4 months that can favor the insertion of implants with good predictability and primary stability than the PRF.

Since the resorption rate has been found to be highest in the first 6 months of tooth extraction and continues at an average of 0.5-1% per year throughout life [3,8,19]. It has been reported also that there is a reduction in the width of the ridge between 2.6 mm and 4.6 mm and height in the range of 0.3-3.9 mm. when MPM is used as a graft [20]. This signifies that the MPM will be the preferable material for grafting for patients who will not prefer surgery (autograft) or allografts because of their high osteogenic potentials from its osteoblastic activity and also because harvest of the graft does not involve any radical surgical procedure.

MPM could also be the best platelets concentrate regenerative material for replacing hard buccal bone in the esthetic zone especially when immediate implants are indicated. Huynh-Ba and colleagues (2010), reported that post-extraction alveolar resorption is significantly more on the buccal surfaces of the alveolar bone than the labial surfaces. In their study, they observed that it may be due to the labial anatomy of the alveolar bone which is thin and knife-edged. They also found out that the thin buccal cortical bone undergoes average resorption up to 0.8 mm in the anterior teeth and 1.1 mm in the premolar sites [17] leading to unfavorable conditions for implant placement. As such in replacing bone loss or compensating bone loss in the esthetic zone, MPM could be the best option because it has the potential of forming a harder and matured bone within 4 months that can favor the insertion of implants with good predictability and primary stability.

Conclusion

It can be concluded from this study that the use of bone filler combined with fibrin, platelets and leukocytes in the form of MPM shows a better histologic evidence of hard bone formation as evidenced by its high osteoblastic activity and maturation than when PRF is used as the sole filling material for the extraction socket after 4 months.

Conflicts of interest

Authors declared no conflict of interests.

Bibliography

1. Ziccardi VB and Buchbinder D. "Guided tissue regeneration in dentistry". *The New York State Dental Journal* 62.10(1996): 48-51.
2. Kumar P, et al. *Journal of Pharmacy & Bio allied Sciences*. 5(2013): S125-S127.

3. Chandna S., et al. "Socket augmentation". *Journal of the International Clinical Dental Research Organization* 7(2015): 73-80.
4. Meloni SM., et al. "Postextraction socket preservation using epithelial connective tissue graft vs porcine collagen matrix. 1-year results of a randomised controlled trial". *European Journal of Oral Implantology* 8.1(2015): 39-48.
5. Artzi Z., et al. "Porous bovine bone mineral in healing of human extraction sockets. Part 1: Histomorphometric evaluations at 9 months". *Journal of Periodontology* 71.6(2000): 1015-1023.
6. Marcus SE., et al. "Tooth retention and tooth loss in the permanent dentition of adults: United States, 1988-1991". *Journal of Dental Research* 75(1996): 684-695.
7. Mecall RA and Rosenfeld AL. "Influence of residual ridge resorption patterns on implant fixture placement and tooth position. 1". *International Journal of Periodontics and Restorative Dentistry* 11.1(1991): 8-23.3.
8. Araújo MG and Lindhe J. "Dimensional ridge alterations following tooth extraction. An experimental study in the dog". *Journal of Clinical Periodontology* 32.2(2005):212-218.
9. Lekovic V., et al. "Preservation of alveolar bone in extraction sockets using bioabsorbable membranes". *Journal of Periodontology* 69.9(1998): 1044-1049.
10. Irinakis T. "Rationale for Socket Preservation after Extraction of a Single-Rooted Tooth when Planning for Future Implant Placement". *Journal of the Canadian Dental Association* 72.10(2006): 917-922.
11. Gupta V., et al. "Regenerative potential of platelet rich fibrin in dentistry: literature review". *Asian Journal of Oral Health and Allied Sciences* 1.1(2011): 22-28.
12. David M., et al. "Classification of platelet concentrates: from pure platelet-rich plasma (P-PRP) to leucocyte- and platelet-rich fibrin (L-PRF)". *Trends in Biotechnology* 27.3(2009): 158-167.
13. Choukroun J., et al. "Une opportunit e en paro-implantologie : le PRF". *Implantodontie* 42(2000): 55-62.
14. Choukroun J., et al. "Platelet rich fibrin (PRF): A second-generation platelet concentrate. Part IV: clinical effects on tissue healing". *Oral surgery oral medicine oral pathology oral radiology & endodontics*. 101.3(2006):56-60.
15. Mazzoni L and P eriss e J. "Apports de la microscopie  lectronique   balayage pour la Matrice Plasmatique Min ralis e". *La Lettre de la Stomatologie* 51(2011).
16. Kim JH., et al. "The use of biodegradable PLGA nanoparticles to mediate SOX9 gene delivery in human mesenchymal stem cells (hMSCs) and induce chondrogenesis". *Biomaterials* 32.1(2011): 268-278.
17. H ammerle CH., et al. "Osteology Consensus Group 2011. Evidence-based knowledge on the biology and treatment of extraction sockets". *Clinical Oral Implants Research* 23.5(2012): 80-82.
18. Ashman A. "Postextraction ridge preservation using a synthetic alloplast". *Implant Dent* 9.2(2000): 168-176.
19. Ten Heggeler JM., et al. "Effect of socket preservation therapies following tooth extraction in non-molar regions in humans: A systematic review". *Clinical Oral Implants Research* 22.8(2011): 779-788.
20. Huynh-Ba G., et al. "Analysis of the socket bone wall dimensions in the upper maxilla in relation to immediate implant placement". *Clinical Oral Implants Research* 21.1(2010): 37-42.

Volume 5 Issue 1 September 2016

  All rights reserved by Ayoub AH., et al.