Platelet rich fibrin–A Healing biomaterial

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Introduction

The development of bioactive surgical additives, which are being used to regulate the inflammation and increase the speed of healing process, is one of the great challenges in clinical research. In this sense, healing is a complex process, which involves cellular organization, chemical signals, and the extracellular matrix for tissue repair.[1,2] The understanding of healing process is still incomplete, but it is well known that platelets play an important role in both hemostasis and wound healing processes.[3] Platelets' regenerative potential was introduced in the 70's, when it was observed that they contain growth factors that are responsible for increase collagen production, cell mitosis, blood vessels growth, recruitment of other cells that migrate to the site of injury, and cell differentiation induction, among others.[4,5]

PRF consists of an autologous leukocyte-platelet-rich fibrin matrix[6], composed of a tetra molecular structure, with cytokines, platelets, cytokines, and stem cells within it[7,8], which acts as a biodegradable scaffold[9] that favors the development of microvascularization and is able to guide epithelial cell migration to its surface.[10] Also, PRF may serve as a vehicle in carrying cells involved in tissue regeneration and seems to have a sustained release of growth factors in a period between 1 and 4 weeks, stimulating the environment for wound healing in a significant amount of time.[11-13] It has a complex architecture of strong fibrin matrix with favorable mechanical properties and is slowly remodeled, similar to blood clot. Some studies have demonstrated that PRF is a healing biomaterial with a great potential for bone and soft tissue regeneration, without inflammatory reactions and may be used alone or in combination with bone grafts, promoting hemostasis, bone growth, and maturation.[14-17] This autologous matrix demonstrated in in vitro studies a great potential to increase cell attachment and a stimulation to proliferate and differentiate osteoblasts. Dohan et al.[18] stated that PRF has immunological and antibacterial properties, may lead to leukocyte degranulation, and has some cytokines that may induce angiogenesis and pro/anti-inflammatory reactions.

Preparation of PRF

The PRF preparation protocol is very simple and armamentarium required is same as that of PRP. Around 5 ml of whole venous blood is collected in each of the two sterile vacutainer tubes of 6 ml capacity without anticoagulant. The vacutainer tubes are then placed in a centrifugal machine at 3000 revolutions per minute (rpm) for 10 minutes, after which it settles into the following layers: red lower fraction containing red blood cells, upper straw coloured cellular plasma and the middle fraction containing the fibrin clot. The upper straw coloured layer is then removed and middle fraction is collected, 2 mm below lower dividing line, which is the PRF. The mechanism which is followed here is that, fibrinogen which is initially concentrated in the high part of the tube, combines with the circulating thrombin due to centrifugation, to form fibrin. A fibrin clot is then obtained in the middle of the tube, just between the red corpuscles at the bottom and acellular plasma at top. Platelets are trapped massively in the fibrin meshes. The success of this technique entirely depends on the speed of blood collection and transfer to the centrifuge. In fact, without anticoagulant, the blood sample starts to coagulate almost immediately upon contact with the tube glass, and it does take a minimum of few minutes of centrifugation to concentrate fibrinogen in the middle and upper part of the tube. Quick handling is the only way to obtain a clinically usable PRF clot.PRF protocol makes it possible to collect a fibrin clot charged with serum and platelets. By driving out the fluids trapped in the fibrin matrix, practitioners can obtain very resistant autologous fibrin membranes.PRF is also called Choukroun's PRF apart from other similar concentrates such as Vivostat PRF and fibrin PRF.

Advantagesof PRF

Numerous studies showed that LPRF has the property of the new bone formation.[20] The use of PRF has reduced healing time by promoting optimum bone regeneration.

In surgical procedures, PRF could serve as a resorbable membrane for guided bone regeneration (GBR)[21], preventing the migration of non-desirable cells into bone defect and providing a space that allows the immigration of osteogenic and angiogenic cells permitting the underlying blood clot to mineralize[22]; moreover, a normal PRF membrane has a rapid degradability (1–2 weeks).[23]

PRF membrane helps in wound healing, protecting the surgical site promoting soft tissue repair; when mixed with bone graft, it may act as a “biological connector”, which attracts

stem cell, favors the migration of osteoprogenitor cells to the center of the graft, and provides a neo-angiogenesis.

In addition, PRF may act as a biologic adhesive to hold the particles together, facilitating the manipulation of the bone grafts.[24-26]

Ross *et al*. in 1974 introduced the regenerative potential of platelets by describing growth factors from platelets.[27] Platelet-rich fibrin (PRF) is a natural fibrin matrix which contains platelet cytokines, growth factors, and stem cells that serves as a resorbable membrane which promotes wound healing and regeneration of periodontal tissues.[28]

Whitman et al. in 1997 concluded that PRF-enhanced osteoprogenitor cells in the host bone and bone graft.[29]Choukroun et al. in 2001 used PRF in cases of implants to induce bone regeneration.[30] PRF is an autologous preparation, which is easy to prepare and does not require any chemical manipulation of the blood.[31]

PRF consists of fibrin matrix which is incorporated by platelets, leukocytes, cytokines and circulating stem cells.[32] Osteoblasts, gingival fibroblasts, and periodontal ligament cells are stimulated by PRF. PRF acts as an ideal matrix for endothelial cell and fibroblast migration.[33] PRF acts as an immune regulator due to presence of leukocytes and immune cytokines like IL 1β, IL 6, IL 4 and TNF α.[34] TGF-beta and PDGF induce collagen production to improve wound strength which promotes healing.[35] Fibrin matrix directs the wound coverage. PRF aids in trapping circulating stem cells and these are brought to the wound site.[36]

Disadvantages of PRF

The disadvantage of platelet rich fibrin is the requirement of an autologous blood source that contains various immune cells and highly antigenic plasmatic molecules in the fibrin matrix. These cells' presence makes platelet rich fibrin donor-specific, therefore limiting its use as an allogenic graft tissue.[8] Additionally, the required use of autologous blood restricts the amount of platelet rich fibrin available, as a low quantity is produced; thus, it can only be used in limited amounts.[8]

Applications of PRF in dentistry

PRF-based membranes are used for masking the alveolar ridge augmentation side in several in vivo studies. L-PRF is a new platelet concentrate used with a great success in a number of surgical procedures to optimize the wound healing.

Tooth extraction has various adverse effects such as pain, bleeding, swelling, infection etc. Wound healing in the tooth extraction is characterized by bone loss as a natural process. Furthermore; extraction will result recession around adjacent teeth and hinders the functional and esthetic prosthetic rehabilitation. PRF have been shown to play an important role in tissue healing with the releasing growth factors from alpha granules, regulate cellular events such as cell adhesion, migration, proliferation, differentiation and extracellular matrix deposition. Major changes occurred within the first year following extraction, but a major part of bone resorption takes place only within 3 months.[37,38]

PRF can also be used in regenerative periodontal therapy to enhance hard and soft tissue wound healing and promote periodontal tissue regeneration. Various studies have shown the favorable benefit of using PRF as an adjunct to traditional periodontal surgical techniques. These studies all exhibit improved clinical outcomes regarding key clinical parameters such as clinical attachment level and pocket depth with the use of PRF when compared to conventional techniques applied alone.

The rationale behind this benefit is believed to lie in the differentiation and proliferation inducing abilities of PRF. The rich source of bioactive cells within PRF itself stimulate the local environment and regulate the inflammation process, thereby enhancing periodontal wound healing and reducing postoperative discomfort. In addition to these benefits the PRF sample also inherently supplies growth factors, releasing them slowly into the wound for 7–14 days. Other obvious benefits include graft stabilization. Furthermore, a possible antimicrobial effect of L-PRF is also present.[40]

The performance of PRF in different periodontal surgery indications was measured and PRF was found to perform superiorly when compared to conventional perio-plastic surgeries applied alone. Its use in intra-bony defects and furcation defects have proved beneficial in reducing pocket depth values, clinical attachment level gains and bone fill percentages. Improved outcomes in intrabony defects were obtained when used alone or in conjunction with other biomaterials. In furcation defects also, traditional flap surgeries tended to perform better when complemented with PRF. Coronally Advanced Flap (CAF) procedures showed

improved results when accompanied with either CTGs or PRF membranes. Compared to each other however, these two materials seemed to perform similarly. Therefore, it can only be said that PRF can be considered a suitable alternative to CTGs in periodontal plastic surgery.[41,42]

Implant rehabilitation success is highly related with sufficient bone volume and density. The posterior maxilla represents a challenging and unique area for successful dental implant rehabilitation because of its relatively deficient bone volume and poor bone quality caused by alveolar bone resorption and maxillary sinus pneumatization. Rehabilitation of posterior maxillary bone volume has been successes by different procedures, such as Le Fort I osteotomies, onlay grafts and sinus lifts.[43,44] Maxillary sinus floor elevation is considered one of the most successful procedures that can be performed using different grafting materials, such as autogenous, xenograft, allograft, alloplast and PRF.[45,46]

Autogenous bone with osteogenic, osteoinductive and osteoconductive properties is still considered to be the gold standard. However, grafting with autogenous bone is associated with donor site morbidity, extended duration of surgical procedures and the volume of bone graft harvested may be insufficient for the requirements. Biomaterials, thus, are promising substitutes for autogenous bone grafts in maxillary sinus augmentation. Osteoconductive properties of these biomaterials have been shown in clinical studies with satisfactory clinical outcomes.On the other hand, these bone graft materials demonstrate lack of osteogenic and osteoinductive potential with distinct osteogenic capacity and bone formation. Moreover, some disadvantages, mainly related to a limited availability, prolonged healing time and impact on host responses can appear when using these bone substitutes. To overcome these problems, new substances with osteoinductive properties, such as platelet-rich fibrin (PRF) was recently introduced as replacement or additional materials in sinus augmentation procedures.[47]

Platelet concentrates have been used to accelerate bone generation and improve healing by releasing growth factors such as transforming growth factor β1 and β2, platelet-derived growth factor and vascular endothelial growth factor, which are able to induce angiogenesis and activate cell proliferation.

In the literature there are some different application techniques for PRF in the sinus augmentation such as PRF as a sole grafting material, PRF with allografts or PRF with

xenografts. All of these techniques have variable clinical, radiographic and histologic and histomorphometric outcomes.

Mazor et al.[48] and Simonpieri et al.[49] performed sinus lift by using lateral approach and PRF was used as a sole grafting material and implants were applied immediately to serve as tent pegs. During the healing period there were no complications. A 100% survival rate was observed in total of 57 sinus lift procedures and 110 implants during the follow-up period (2 years). Radiographic examination was performed by CT scan or panoramic radiographs about 6 months after the sinus augmentation to examine the bone volume, where the average bone gain was 9.8 mm. Histologic and histomorphometric examination accomplished by Mazor et al. showed that dense collagen matrix, easily identified osteocytes and osteoblasts in the lacunae and well-organized and vital bone with structured trabeculae with more than 30% bone matrix.

Choukroun et al.[50] performed sinus augmentation with PRF in combination with demineralized freeze-dried bone allograft (DFDBA). They found the rate of vital bone/inert bone 20% both in test and control group but with a reduced healing time at PRF group.

Marginal bone loss is an inevitable process which starts immediately following implant placement. There have been done plenty of studies since decades to minimize it. Previous studies about preserving bone around implants, has focused on soft tissue thickness and it was hypothesized, adequate soft tissue volume around implants has a positive effect in preserving marginal bone and PRF is perfect material to augment soft tissue. We know PRF is a good autologous material to enhance soft tissue healing with its growth factors including VEGF, PRGF, etc. However researches about PRF usage to augment hard tissue have contradictory results and there is need to do further detailed randomized controlled clinical studies to know about the effect of PRF preserving marginal bone.[51]

In Endodontics, PRF was used in infected necrotic immature tooth for pulpal regeneration and revitalization and thus concluded that PRF acts as a scaffolding material.[52]

Successful healing and apexification with the combined use of MTA as an apical barrier and autologous platelet-rich fibrin membrane as an internal matrix were reported by Rudagi K. and B. Rugadi. [33].

Additionally, PRF enhanced dental pulp cell proliferation, upregulation in alkaline phosphatase activity, and increased osteoprotegerin expression in a time-dependent fashion.[53]Pulpotomy in young permanent teeth using PRF has been reported affirmative .[54]

Conclusion

Thus, with this article we can conclude that the new and recent generation of platelet concentrate-PRF, would be a good friend to dentists in the near future. The clinical experience also confirms that PRF can be considered a healing biomaterial, as it features all the necessary parameters permitting optimal wound healing. It already has a list of intraoral applications, and numerous extraoral applications can also be imagined.PRF have safe and promising results when used as a sole material or an adjunct to other biomaterials

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