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Review Article

Exploring the Role of Platelet Derived Concentrates in Accelerated Orthodontics - A Narrative Review

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Abstract

Devising new approaches to hasten the regenerative process, thereby, waning the duration of wound healing persists to be one of prime goals of clinical research. Platelets play a crucial role in these regenerative procedures, as they act as reservoirs of cytokines and growth factors. A significant component of the orthodontic tooth movement encompasses a similar process as it essentially involves bone remodeling comprising of alternate cycles of bone resorption and deposition. The past decades have witnessed various attempts to accelerate the rate of orthodontic movement by versatile techniques leading to a decrease in the treatment time, a highly desired outcome by the patient as well as the orthodontist. The following review attempts to summarize existing literature concerning the application of platelet derived concentrates in dentistry and their potential role in orthodontics. The most commonly used platelet derived concentrates i.e Platelet Rich Plasma (PRP), Plasma rich in growth factors (PRGF) and the Platelet rich Fibrin (PRF) have been described and compared to aid in future opportunities of clinical research.

Keywords: Platelet-rich Plasma; Platelet-rich Fibrin; Orthodontics; Regenerative Medicine

Introduction/Description

Long treatment duration persists to be one the prime concerns perturbing the patient before treatment commencement. More often than not, the long treatment duration associated becomes a deterrent discouraging the patient from undergoing treatment. Consequentially, orthodontic tooth movement and the associated biological reaction persist to be one the prime research domains.

Orthodontic tooth movement involves modeling-remodeling of the alveolar bone caused by a series of inflammatory mediators released on the application of an external force [1-3]. The process of remodeling is essentially a healing process initiated by the formation of a blood clot, followed by a proliferative stage comprising of new epithelium, blood vessel and granulation tissue formation ultimately leading to deposition of collagen and bone [4]. The clot formation involves platelet aggregation and adherence favoring the formation of thrombin and fibrin.

Many surgical and non-surgical approaches have been carried out over the years by researchers to enhance the orthodontic tooth movement while preserving the bone physiology. The surgical category is based on the principle of the Regional Acceleratory Phenomenon described by Frost [5], where surgical irritation of the bone releases an inflammatory cascade leading to osteoclastogenesis. Subsequently, alveolar bone resorption ensues and results in a decrease in the thickness and weight of the alveolar bone, thus, producing accelerated tooth movement [6]. A majority of the surgical procedures such as alveolar decortication, distraction of the periodontal ligament or the dento-alveolus [7] follow this principle and produce an insult to the bony tissue to accelerate tooth movement. However, these interventions were deemed to be traumatic and thus, less invasive and non invasive procedures, thus, such as injections of prostaglandins [8,9], osteocalcin [9,10], active form of vitamin D [8,10], resonance vibration [11], photobiomodulation including low level laser therapy [12,13], LED and ultrasound

waves [14] were introduced. Though some of the pharmacological substances did stimulate the rate of tooth movement, one could not overlook the undesirable side effects of local pain and discomfort and during a shorter duration of action [6].

The concepts of regenerative surgery where surgical annihilation techniques are often combined with regenerative methods to attain ideal healing have been widely used in various aspects of dentistry such as intrabony defects, gingival recession, sinus lifting and alveolar filling post extraction [15,16]. The bioactive properties of platelets in the healing process have enabled them to be used as a therapeutic and non-invasive adjunct for these various processes. Thus, the application of autologous platelet concentrates in the field of orthodontics may prove to be promising and thus warrants the need for extensive research.

Platelet concentrates and their evolution

Platelet concentrates are products derived from the centrifugation of blood concentrating platelets, leukocytes and fibrin to convert them into a clinically useful form. The efficiency of the concentrate depends on the platelet concentration, number and type of leukocytes in the fibrin membrane and the subsequent release of bioactive molecules from the site of the clot triggering the regenerative process [17]. The bioactive molecules released include growth factors such as platelet-derived growth factor (PDGF), epidermal growth factor (EGF), vascular endothelial growth factor (VEGF), insulin-like growth factor (IGF-I), hepatocyte growth factor (HGF), transforming growth factor-β (TGF-β) and cytokines such as interleukin-1 (IL-1), interleukin-4 (IL-4), interleukin-6 (IL-6), interleukin-10 (IL-10) and chemotactic molecules such as exotoxin and chemokine ligand-5 (CCL- 5) [18]. The initial protocols involved the formation of first-generation platelet concentrates i.e. the platelet-rich plasma (PRP) and plasma rich in growth factors (PRGF). The expansion in knowledge about the biological features of the concentrates led to the development of the second-generation concentrates i.e. leukocyte-platelet-rich fibrin (L-PRF/PRF). The evolution is based on the principle of addition of no anticoagulants and no manipulation of the blood sample to obtain the concentrates [19]. Further advances in the field of platelet concentrates include the development of CGF (Concentrated Growth Factors), A-PRF (Advanced PRF) and i-PRF (injectable form of PRF). Concentrated Growth Factors (CGF) were introduced by Sacco [20] in 2006 where the preparation of the concentrates is similar to PRF but at a different speed. This allowed the separation and formation of a fibrin matrix which was much larger, denser and richer in growth factors. Choukroun [21] further modified the L-PRF and introduced a concentrate called the A-PRF in an attempt to incorporate monocytes in the PRF and produce faster soft tissue growth, more growth factors and cytokines release. An injectable form of PRF called the i-PRF was described by Mourão., *et al.* [22] in 2015, wherein he obtained an orange color fluid instead of the matrix by a shorter centrifugation of blood. This fluid could either be mixed with the graft or injected separately.

However, as these advances are fairly new, no conclusive long term or controlled trials in humans or animals have been carried out to prove their efficiency over the conventional PRP and PRF. Thus, researchers have cautioned against the use of these products till further conclusive results have been attained.

Platelet rich plasma

Platelet rich plasma or PRP was introduced in dentistry by Robert Marx [23] to enhance the radiographic maturation rate of bone graft in mandibular reconstructive procedures. PRP is an autologous concentration of human platelets in a small volume of plasma comprising of a high concentration of platelets with fundamental growth factors actively secreted by them during wound healing [24]. Platelet counts in PRP are approximately 1,000,000/µl compared the normal range of 150,000/ µl to 350,000/µl [25].

Composition [26]

Platelet rich plasma (PRP) comprises of:

High concentration of platelets which release the following growth factors:

- Three isomers of platelet-derived GF (PDGFaa, PDGFbb, and PDGFab)
- Transforming GFs-b (TGFb1 and TGFb2),
- Vascular endothelial GF (VEGF)
- Epidermal GF (EGF).

Small volume of plasma containing

- Cytokines, Interleukins and Tumour Necrotic Factor
- Cell adhesion proteins – Fibrin, Fibronectin and Vitronectrin
- Proteases and anti proteases.

Method of preparation [27]

According to a study by Raja and Naidu [27], PRP can be prepared by two techniques differing in their technical aspects.

General purpose separators

General-purpose cell separators such as ELMD-500 (Medtronic Electromedic, Auto Transfusion System, Parker, CO, USA) requires a large quantity of blood (approximately 450 ml) and are usually operated in an hospital setting.

Platelet-concentrating cell separators

These separators require a smaller quantity of blood (10 - 50 mL) to produce PRP and thus are more commonly used in dentistry e.g. Harvest SmartPrep Platelet Concentrate System (HSPCS; Harvest Technologies, Plymouth, MA, USA) and the 3i Platelet Concentrate Collection System (3i PCCS; 3i Implant Innovations, Palm Beach Gardens, FL, USA).

Robert Max originally produced PRP with a double centrifugation technique as a single spin would not produce a true PRP but a mixture of platelet poor plasma and platelet rich plasma with very low platelet counts [24]. The processing of platelet rich plasma essentially differs in the type of anticoagulant and duration of centrifugation in the various platelet-concentrating systems.

PRP is usually obtained in an injectable form but the addition of bovine thrombin and calcium chloride just before application converts it into a gel form. Calcium chloride is added to nullify the effect of the citrate anticoagulant used earlier and the thrombin aids in activation of fibrinogen required to form fibrin [28]. However, in the field of orthodontics, an injectable form without the use of calcium chloride and thrombin has been advocated to have a longer lasting effect of the product [9] (Figure 1).

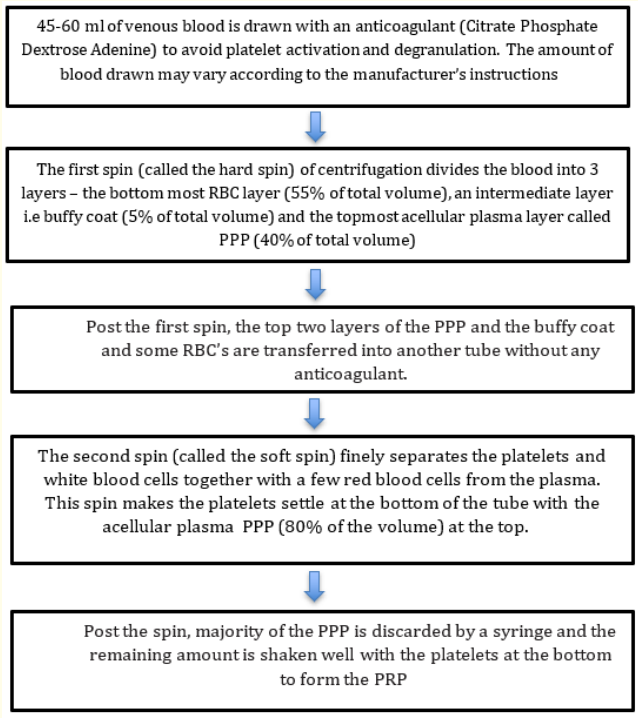

Figure 1: Steps in preparation of PRP.

Applications

PRP and Accelerated orthodontics

A study by Graziani., *et al.* [29] has stated that the role of PRP in bone metabolism is highly dependent of the concentration of the platelets in the product. Rashid., *et al.* [30] and Güleç., *et al.* [31] in their animal studies have found significant results in the role of PRP in accelerating the rate of orthodontic tooth movement. However, there still exists a huge lacuna as there is a paucity of any conclusive human studies concerning the same.

PRP for alveolar bone grafting in cleft patients

The role of PRP in cleft patients is based on the release of the growth factors by the platelets to aid in bone graft maturation. In a study by Gupta., *et al.* [32], secondary alveolar bone grafting with the use of PRP showed significantly higher bone density up to 6-months post-surgery. Over the next few years, various studies [33,34] have elaborated the role of PRP used with an autologous bone graft or other bone substitutes and have demonstrated the positive effect of PRP on the soft tissue by increasing the cell tropism and accelerating the healing process.

Drawbacks of PRP

The PRP concentrate does not promote bacterial proliferation, as it is similar to a natural clotting process. The growth factors released by the platelets in PRP are only trans-membranous and not mutagenic, thus they simulate the natural healing process without causing any tumor [2].

Plasma rich in growth factors

To overcome the drawback associated with PRP such as the low handling efficiency or the addition of thrombin [35], PRGF (Plasma

rich in growth factors) was developed by Anitua E [36] by introducing a modification in the preparation protocol. The thrombin was replaced with calcium for clotting. Anitua E [36] describes PRGF as an autologous platelet enriched plasma without leukocytes. PRGF contains platelet and plasma growth factors along with plasma proteins such as fibrin, vitronectin and fibronectin involved in the wound healing process.

Composition [20]

The composition of PRGF is essentially similar to PRP i.e.

Growth factors such as:

- Platelet-derived growth factors
- Transforming GFs-b (TGFb1 and TGFb2)
- Vascular endothelial GF (VEGF)
- Epidermal GF (EGF)
- Insulin like growth factor (IGF-1).

Plasma containing

- Cytokines, Interleukins and Tumor Necrotic Factor
- Cell adhesion proteins - Fibrin, Fibronectin and Vitronectin
- Proteases and anti proteases.

Method of preparation [33]

The preparation of PRGF also requires a small volume of blood, which is adapted to each case ranging from 5-80 cm^3. Sodium citrate and calcium chloride are added as anticoagulants (Figure 2).

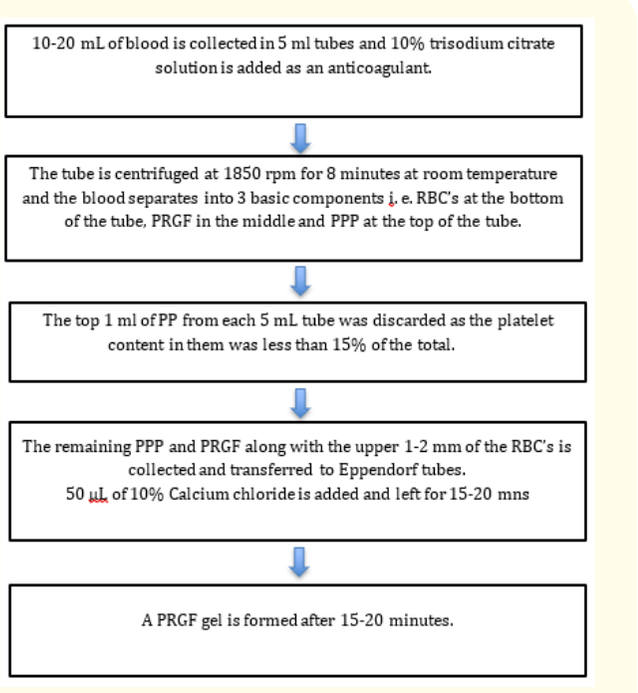

Figure 2: Steps in preparation of PRGF.

Applications

Studies of the application of PRGF in orthodontics and accelerated orthodontics are scarce. However, PRGF has been used in the other fields of dentistry for their regenerative properties such as [37].

- Improving osseointegration by soaking the implant in PRGF before placement.
- Support bone regeneration by via chemotactic and mitogenic effects on preosteoblastic and osteoblastic cells

Drawbacks of PRGF [15]

A high concentration of thrombin added during the preparation of PRGF allows thickening of the polymers of fibrin resulting in formation of rigid meshwork favoring the sealing of the biological tissues but impairing cytokine release and cellular migration.

Platelet rich fibrin

Platelet-rich fibrin (PRF) first described by Choukroun., *et al.* [38] is essentially a platelet concentrate containing platelets and growth factors in the form of fibrin membranes. It is prepared from autologous blood and is free of any anticoagulant and other artificial biochemical modifications.

PRF presents itself with several advantages compared to PRP and PRGF [15,26,39]:

- Easier preparation without any chemical manipulation of the blood making it an autologous preparation
- The clot produced forms a flexible matrix favoring the entrapment of cytokines and cells
- Its preparation is a simplified technique with minimum blood manipulation.
- No addition of any external agent such as thrombin is required.
- It comprises of a natural fibrin framework composed of growth factors that have activity for a relatively longer period.
- It is an economical and quick option compared to recombinant growth factors.

Composition

PRF is essentially an autologous leukocyte-platelet-rich fibrin matrix. It is a tetra molecular structure containing platelets, cytokine and stem cells that act as a biodegradable scaffold [17,36].

PRF forms a natural blood clot with a dense fibrin network. This fibrin network contains cytokines, leukocytes, structural glycoproteins and growth factors such as platelet-derived growth factor, vascular endothelial growth factor, transforming growth factor b1, and glycoproteins such as thrombospondin-1 [37].

As the preparation of PRF does not involve the addition of any anticoagulant, as soon as the blood comes in contact with the glass surface of the tube, it begins to coagulate. The contact with a silica surface is required to activate the clot polymerization process. In addition, the silica particles unlike the bovine thrombin used for PRP preparation, are not cytotoxic [26].

Method of preparation [37]

The method of preparation involves centrifugation of blood without addition of any anticoagulant or bovine thrombin. The blood sample is drawn and immediately centrifuged to obtain the product (Figure 3).

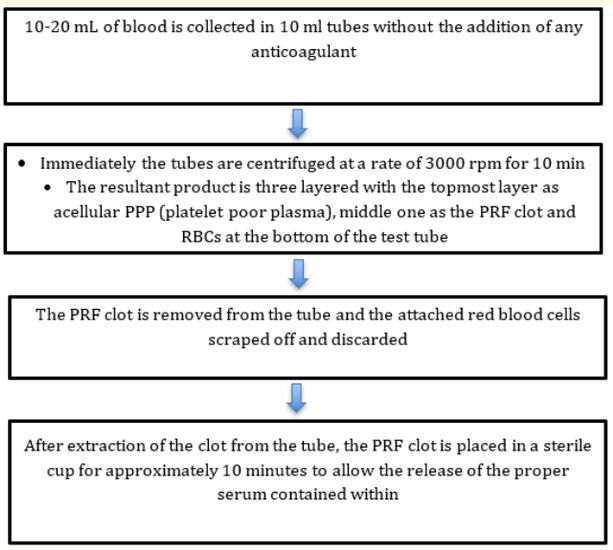

Figure 3: Steps in preparation of PRF.

Applications

Accelerated orthodontics

Tehranchi., *et al.* [39] conducted a human pilot study evaluating the effect of PRF (placed in extraction sockets) on orthodontic tooth movement (OTM). They found a possible positive efficacy of PRF application in the extraction socket for acceleration of OTM. However, there exists a paucity of conclusive human studies to ascertain the same results.

- PRF aids in achieving a reduction in probing depth in periodontal defects along with acting as an adjunct for palatal wound healing after harvesting a soft tissue graft [40].
- PRF can act as a potential scaffold in pulp revascularization procedures of necrotic immature permanent tooth as it is rich in growth factors [41].
- It can be used for preservation of alveolar ridge height and bone regeneration around immediate implants [42].

Drawbacks [43]

- The success of the PRF depends on the handling during the blood collection time and its transference for further process.
- Need of using a glass-coated tube for clot polymerization.

S No	Growth factor	Source	Biologic action
1	Platelet-derived growth factor (PDGF) [44]	Alpha granules (Platelets), Macrophages, Endothelial cells, Monocytes, fibroblasts	Favor angiogenesis and collagen synthesis Increase the rate of proliferation of stem cells.
2	Transforming Growth Factor-ß. [45]	Platelets, T lymphocytes, neutrophils and bone extracts	Chemotaxis and mitogenesis of undifferentiated cells to the place of repair activating fibroblasts, osteoblasts, and chondroblasts proliferation Inhibitory effect on osteoclasts
3	EGF (Epidermal Growth Factor) [46]	Epithelial cells	Promotes chemotaxis and mitogenesis of epithelial, mesenchymal cells and fibroblasts also inducing tissue regeneration.
4	IGF 1 – (Insulin like growth factor) [47,48]	Osteoblasts, macrophages, monocytes, chondrocytes	Mediates growth, differentiation, and cellular transforming and stimulates osteoblasts. It is also involved in keratinocytes migration and wound healing
5	VEGF (Vascular endothelial growth factor) [49]	Macrophages, Platelets, Keratinocytes	Signaling protein stimulating chemotaxis and endothelial cell proliferation with specificity for the vascular endothelial cells Regulates vascular permeability
6	HGF (Hepatocyte growth factor) [50]	Macrophages, Fibroblasts	Regulates the migration and cellular morphogenesis

Table 1: Growth factors released during wound healing and their biological actions.

Platelet Concentrate	PRP	PRGF	PRF
Generation	1st	1st	2nd
Coagulation product added	Citrate phosphate dextrose adenine followed by thrombin and calcium chloride	Sodium citrate and Calcium chloride	None
Speed of centrifugation	Slow	Very slow	Fast
Amount obtainable	Moderate	Poor	Good
Cost	High	High	Low
Fibrin formation	None	Yes	Yes
Fibrin morphology		Tetramolecular	Tri molecular
Leukocytes		Nil	65%
Growth factors	✓	✓✓	✓✓✓
Presentation form	Liquid/ Gel	Liquid Clot Supernatant Fibrin membrane	Plugs Exudate Fibrin membrane

Table 2: Comparison between the first and second-generation platelet concentrates [19,38].

Conclusion

Several *in vitro* and *in vivo* studies in the past have evaluated the efficiency of platelet concentrates and have established safe results for their use in humans. As a result, a wide of clinical applications have been established in dentistry where the concentrates are highly beneficial.

However, in the field of orthodontics there still exists a huge lacuna of the role of these products for accelerating tooth movement, as there is a paucity of any conclusive human studies concerning the same.

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Conflict of Interest

The authors declare that they have no conflict of interest.

Availability of Data and Materials

Not applicable.

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