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Platelets in Focus

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Abstract

Platelets have long been recognized as vital molecules in the process of hemostasis yet have further been shown to be resourceful structures for blood vessel formation, inflammatory responses, innate immunity and especially wound healing. In this context, the applicability of platelet concentrates as adjuvants to the tissue repair process is evident, especially Platelet-Rich Fibrin (PRF) associated. Despite controversies, studies emphasize a satisfactory action of PRF in wound healing processes, as well as a facilitating bone repair agent. Research is encouraging for the use of platelet concentrates as a supplement for tumor immune therapies and antibacterial tools. However, the multiplicity of existing preparation protocols based on time and velocity variations still present a difficulty in establishing specific clinical protocols. Thus, the purpose of this mini-review note was to provide an overview of platelet properties, as well as the advances of PRF matrices as tissue repair facilitating agents.

Keywords: Blood platelets, Biological factors, Platelet-rich fibrin, Wound healing, Regeneration

Introduction

Blood platelets have long been recognized as noteworthy mediators of hemostasis guided by the release of bioactive molecules stored in specific organelles. This regulated exocytosis pathway is maintained by a complex membranous system responsible for the interplay between the cytoplasm and the surrounding microenvironment [1]. Over the years, a concept of extensive versatility has been assigned to platelets, apart from hemostatic activities, as indispensable structures for numerous physiological responses including angiogenesis [2], inflammation [3], innate immunity [4] and wound healing [1].

Platelets are anucleate cell fragments of 1 to 4μm in diameter released from Megakaryocytes (MKs), located in the bone marrow, through a multiple stage biological mechanism at a 100 billion/day rate [6]. Circulating blood platelets exhibit a discoid structural configuration with an internal tubular system connected to the platelet surface, which is responsible for regulating secretory mechanisms. Their complex architecture maintained by an exclusive extent of receptors [7] makes platelets highly reactive and assures the efficiency of hemostatic functions in response to vascular ruptures, which combine adhesion of circulating platelets, activation, and aggregation phases [1,8]. More than 300 biologically active substances released from platelets are involved in the tissue repair process [9].

Platelet activation has been characterized as being guided by secretion from three different sections: dense granules, α-granules and lysosomes. The latter consist of a diversified enzyme deposit, whereas dense granules primarily contain small molecules related to platelet aggregation and α-granules store proteins responsible for adhesion and repairing factors [10]. The presence of a sophisticated internal membranous system, composed of specific features, maintains the regulation of the excretory apparatus. The Open Canalicular System (OCS), characterized as an extensive intracellular anastomosing network of fenestrated channels [11], is essential for the proper functioning of platelet activities, such as the uptake and release of substances stored in platelets to the cell exterior. Furthermore, the OCS works as a membrane reserve resource, available for replacement during platelet activation morphological changes, as well as a storage site for platelet membrane receptors [12]. On the other hand, the Dense Tubular System (DTS), a smooth endoplasmic reticulum membrane structure replete of amorphous material, plays a crucial role in platelet activation. It is a site for prostaglandin and thromboxane synthesis [13], storage of calcium and adenylate cyclase (cAMPase) mobilized in metabolic processes [14], as well as platelet protein disulfide isomerase, which regulates the procoagulant activity of tissue factor, thus controlling the initial phase of coagulation [15].

In vascular injury situations with endothelial discontinuity, platelet activation is mediated by receptor interactions, which results in morphological adjustments. Activation of multiple signaling pathways results in the release of the diversified granule contents. During this process, platelets exhibit a contracted structure via actin/myosin interaction mediated by an increased intracellular calcium ion concentration. A notable event in this context consists of the GPIIb/IIIa receptor exposure which leads to plasmatic fibrinogen binding, thus resulting in further changes to platelet structural conformation [16]. Accordingly, the platelets emit pseudopods and change from the original discoid structure to completely spread increasing the surface area, and stimulating the aggregation of other platelets [17]. Concurrently, a fibrin mesh is organized around the developed clot, acting as an encapsulation mechanism which provides additional stability [9].

The regenerative ability of platelets has been increasingly explored, due to the wide range of mediators in the tissue repair process that are correlated to these molecules. The platelets release a wide range of biomolecules, including growth factors, such as Fibroblast Growth Factor-b (FGFb) and Platelet-Derived Growth Factor (PDGF), chemokines, cytokines, proteins and enzymes. These molecules establish a distinct microenvironment conducive to the development of angiogenic properties, cell recruitment,

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proliferation and differentiation activities, as well as acting upon anti-microbial responses [5]. Furthermore, platelets play a significant role in maintaining the balance between cell death and survival, either through secretion of apoptosis mediators or antiapoptotic mechanisms [18].

Accordingly, the concept of platelet concentrates as potential tissue repair facilitator resources has emerged in modern tissue engineering as an extensive growth factor and cytokine source, especially related to Platelet-Rich (PRF) [19]. Scientific evidence demonstrates the extensive applicability of PRF aggregates. *In vitro* studies have shown the functionality of PRF as bioscaffolds and growth factor reservoir [20-23], as well as the ability to stimulate human periosteal [24] and osteoblast cell activity [25,26]. Furthermore, PRF has also been shown to *in vitro* stimulate bone marrow mononuclear cells [27] tendon cell [28] meniscocytes [29], chondrocytes [30], and heighten gingival mesenchymal stem cell differentiation capacity [31].

Studies on animals have shown the relevance of platelet concentrates for fracture healing [32], bone defects [33] and the osseointegration process over implant interfaces [34], as well as for wound healing in diabetic mice [35]. Regarding clinical studies, the PRF matrices provided favorable results for soft tissue repair [36], assistance in bone ridge preservation as a socket filling material [37] and in the closing process of bone exposures associated with osteonecrosis [38]. PRF has also been successfully employed in the repair of periodontal intrabony defects [39,40], and may even contribute to postoperative pain control after mandibular third molar extraction [41]. The use of PRF matrices in implant dentistry techniques also exhibits clinically acceptable results, as it has been shown to facilitate osseointegration [34,42] as well as increase the width of keratinized mucosa around implants [43].

The biological principle of PRF is to obtain a fibrin clot composed of a platelet and cell-rich network, acting as a conductive, inductive, and histogenic autograft. For this purpose, centrifugation protocols separate the formed blood elements into layers according to their different densities [44]. Over the years, preparation protocols based on distinct time periods and Relative Centrifugation Force (RCF) variations, as well as the introduction of the Low Speed Centrifugation Concept (LSCC), have opened the possibility of producing advanced matrices with an enhanced amount of leukocytes, platelets, growth factors and injectable PRF [45,46]. In this context, there exists a remarkable diversity of centrifugation protocols and, consequently, an elevated number of different PRF matrices, which produce a number of difficulties when defining specific clinical protocols for exact procedures. However, prospects are encouraging, in view of the application of platelet concentrates as a supplement for immune therapies [47] and even as a long-term antibacterial tool, even though further investigation is required [48].

In conclusion, blood platelets and platelet aggregates constitute resourceful structures with hemostatic, angiogenic, inflammatory and chemotactic properties that result in vast applicability for tissue repair therapies. Significant advances are observed regarding tumor and antibacterial therapies, although detailed research is necessary.

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