

# Platelet Rich Concentrate: Basic Science and Current Clinical Applications

## FDA CLEARED INDICATIONS FOR USE

The Double Syringe (ACP) System is used to facilitate the safe and rapid preparation of autologous platelet-rich-plasma (PRP) from a small sample of blood at the patient's point of care. The PRP can be mixed with autograft and allograft bone prior to application to an orthopedic surgical site as deemed necessary by the clinical use requirements.

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This article describes indications for use that are not cleared by the FDA.

**Abstract:** Improvements in resuscitation, dissemination of ATLS protocols, and growth of regional and local trauma centers has increased the survivability after severe traumatic injuries. Furthermore, advances in medical management have increased life expectancy and also patients with orthopaedic injuries. While mechanical stabilization has been a hallmark of orthopaedic fracture care, orthobiologics are playing an increasing role in the management of these patients with complex injuries. Platelet-rich concentrate is an autologous concentration of platelets and growth factors, including transforming growth factor-beta (TGF- $\beta$ ), vascular endothelial growth factor (VEGF), and platelet-derived growth factor (PDGF). The enhancement of bone and soft tissue healing by the placement of supraphysiologic concentration of autologous platelets at the site of tissue injury or surgery is supported by basic science and clinical studies. Due to the increased concentration and release of these factors, platelet-rich plasma can potentially enhance the recruitment and proliferation of tenocytes, stem cells, and endothelial cells. A better understanding of platelet function and appropriate clinical use is essential in achieving the desired outcomes of platelet-rich concentrate in orthopaedic clinical applications.

**Key Words:** platelet rich concentrate

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While mechanical solutions have been the mainstay of orthopaedic interventions for musculoskeletal conditions, the search for alternative treatment strategies is currently underway. One broad category of potential therapeutic interventions falls in the realm of orthobiologics. Traditionally, orthobiologics have been defined as bone graft substitutes and resorbables, which have already begun to replace autografts

and allografts. However, orthobiologics encompass a much wider range of products, from macroscopic, large-scale bone graft substitutes to microscopic, cellular growth factors and bone morphogenic proteins. New technology and techniques are constantly being introduced as orthobiologics is one of the largest growing sectors within orthopaedic surgery and medical devices in general. Given all of the new orthobiologic agents readily available to the orthopaedic surgeon, there is very little clinical evidence to provide a foundation for its use. This is especially true for platelet-enriched plasma, also known as platelet-rich plasma (PRP), platelet-rich concentrate (PRC), autogenous platelet gel, or platelet releasate.<sup>1</sup> The goal of this article is to provide background information on PRC and offer potential orthopaedic conditions for its application. By reviewing the basic science and clinical literature, the goal is to provide the orthopaedic surgeon with evidence-based guidelines for the correct use of platelet concentrates.

## BACKGROUND

Since 1990, a greater understanding of wound, soft tissue, and bone-healing has revealed that there are several components within blood constituents [eg, fibrin, fibronectin, vitronectin, platelet-derived growth factor (PDGF), transforming growth factor-beta (TGF- $\beta$ )] that are part of the natural healing process, which can be altered or accelerated by concentrating these factors. These proteins set the stage for tissue healing (histopromotive factors), which includes cellular chemotaxis, proliferation, and differentiation, removal of tissue debris, angiogenesis, and the laying down of extracellular matrix.<sup>2</sup>

Hematoma and clot formation after surgical intervention or trauma initiates the healing cascade. Clot formation is initiated by 1 of 2 pathways: intrinsic and extrinsic. The intrinsic pathway is initiated by damage or alteration to the blood itself, whereas the extrinsic pathway is initiated by contact of the blood with factors that are extraneous to the blood (eg, damaged tissue). Both pathways involve a cascade of events that, while beginning differently, converge during the latter steps of the process. Platelets and the release of their proteins are essential and necessary for either pathway of clot formation.

Platelet activation in response to tissue damage and vascular exposure results in the formation of a platelet plug and blood clot to provide hemostasis and the secretion of biologically active proteins. The composition of this naturally occurring hematoma is 95% red blood cells, 4% platelets, and 1% white blood cells. However, an analysis of platelet-enriched clot reveals dramatic differences in its composition

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The device that is the subject of this manuscript has been cleared through the Premarket Notification 510(k) process/approved through a Premarket Approval application for the rapid preparation of autologous platelet-rich concentrate (PRC) from a small sample of blood at the patient's point of care, but not for the surgical indications that were reported.

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compared to natural clot with 95% platelets (as opposed to 4%), 4% red blood cells (as opposed to 95% red blood cells), and a similar amount of white blood cells.

PRP is defined as a portion of the plasma fraction of autologous blood having a platelet concentration above baseline.<sup>1</sup> As such, PRP contains not only a high level of platelets but also the full complement of clotting factors and secretory proteins. PRC is an autologous concentration of platelets and growth factors that includes TGF- $\beta$ , vascular endothelial growth factor (VEGF), and PDGF. Due to the increased concentration and release of these factors, PRP can potentially enhance the recruitment and proliferation of tenocytes, stem cells, and endothelial cells.

## PLATELET BIOLOGY

Platelets are the end products of megakaryocytes and are formed in bone marrow. They have no nucleus and cannot replicate. Thus, the life span of a platelet is 5 to 9 days. Originally, it was believed that platelets only created a “plug” during initial hemostasis. This plug is created after tissue injury and/or surgical stimuli when platelets become exposed to damaged blood vessels, which places them in direct contact with various extracellular proteins. This interaction causes the platelets to aggregate at the site. This process is called activation. During activation, the alpha granules within platelets fuse with the platelet plasma membrane and release some of their protein contents to the surroundings (degranulation). The alpha granules in platelets contain more than 30 bioactive proteins, many of which have a fundamental role in hemostasis and/or tissue healing.<sup>3</sup> These proteins include PDGF (including  $\alpha\alpha$ ,  $\beta\beta$ ,  $\alpha\beta$  isomers), TGF- $\beta$  (including  $\beta 1$  and  $\beta 2$  isomers), platelet factor 4, interleukin-1, platelet-derived angiogenesis factor, VEGF, epidermal growth factor, platelet-derived endothelial growth factor, epithelial growth factor, insulin-like growth factor, osteocalcin, osteonectin, fibrinogen, vitronectin, fibronectin, and thrombospondin-1.<sup>3</sup> Platelets begin actively secreting these proteins within 10 minutes after clotting, with more than 95% of the presynthesized growth factors secreted within 1 hour.<sup>4</sup> After the initial burst of PRP-related growth factors, the platelets synthesize and secrete additional growth factors for the remaining several days of their life span.<sup>4</sup>

## Platelet-expressed Growth Factors and Cytokines

### Platelet-derived Growth Factor

PDGF is a dimeric protein consisting of 2 subunits:  $\alpha$  and  $\beta$ . Therefore, it can exist in 3 combinations: PDGF- $\alpha\alpha$ , PDGF- $\beta\beta$ , and PDGF- $\alpha\beta$ . The role of each of these isomers in bone and soft tissue healing is not well explored. PDGF is the first growth factor to start nearly all wound healing.<sup>5</sup> When activated, the growth factor attaches to transmembrane receptors on target cells, including osteoblasts and fibroblasts. The main function of PDGF is stimulating cellular replication (mitogenesis). This growth factor increases cell populations of healing cells, including mesenchymal stem cells and osteoprogenitor cells, which are part of the connective tissue-bone healing cellular composite and endothelial cells, causing budding of new capillaries into the wound (angiogenesis).<sup>6</sup>

PDGF also stimulates bone resorption by increasing the number of osteoclasts, which can lead to faster bone remodeling. Furthermore, PDGF activates macrophages, resulting in debridement of the surgical or traumatic site. The macrophage activation then triggers a second source of growth factors released from the host tissues under the influence of the macrophage action. This secondary release of endogenous factors continues the process of repair and bone regeneration.

In vivo, PDGF in concentrations of 20 to 100 ng has been shown to increase ectopic bone formation and alkaline phosphatase activity using demineralized bone matrix as a carrier in rats. PDGF, when locally applied, can also stimulate bone healing in rat osteotomies, as well as in rabbit calvaria defects in combination with barrier membranes. This protein has also been found to be expressed by many cell types during different stages of normal fracture healing.<sup>7</sup>

### Transforming Growth Factor- $\beta$

TGF- $\beta$  is synthesized by many tissues, but bone and platelets are the major source of this cytokine. TGF- $\beta$  is a polypeptide that stimulates the proliferation of osteoblast precursor cells, and it has direct stimulatory effects on bone collagen synthesis. Therefore, TGF- $\beta$  modulates bone matrix synthesis by increasing the number of cells capable of expressing the osteoblast genotype, as well as direct upregulation of osteoblasts. TGF- $\beta$  also decreases bone resorption by inducing apoptosis of osteoclasts.<sup>5</sup>

In addition to osteoblasts, TGF- $\beta$  activates fibroblasts to induce collagen formation, endothelial cells for angiogenesis, chondroprogenitor cells for cartilage, and mesenchymal cells in an effort to increase the population of wound healing cells.

In vivo, however, TGF- $\beta$  alone failed to induce ectopic bone formation in an animal model. The bone-forming properties of TGF- $\beta$  are maximized when combined with a carrier, like demineralized bone matrix, or when placed exogenously into a fracture site.<sup>8,9</sup>

## Associated Cytokines, Growth Factors, and Platelet Proteins

Fibronectin and vitronectin are cell adhesion molecules that help cells move during the proliferation and migration phases seen in bone and cartilage healing. Fibrin contributes to cell mobility in the wound by serving as a scaffold for cell migration and platelet entrapment. Crosslinking occurs as part of the clotting process and ensures a random distribution of platelets and their growth factors throughout the wound. In PRP, the concentration of these expressed proteins is increased, potentiating accelerated bone and soft tissue healing.

### Creation of PRC

Deriving clinically effective platelet concentrate from autologous blood is rooted in specific principles: platelet concentrate ratio, processing technique, quantification of secretory protein concentration, handling and application, and clinical use.<sup>10</sup> Some investigators have suggested that platelet-rich concentrate should achieve a 3- to 5-fold increase in platelet concentration over baseline.<sup>11</sup> However, the dependence of clinical benefit on platelet concentration versus total number of platelets delivered may need to await further

investigation as concentration ratios of less than 2-fold and greater than 8.5-fold have been reported. Because most individuals have a baseline blood platelet count of 200,000/ $\mu$ L, a PRP count of 1,000,000/ $\mu$ L as measured in a standard 6-mL aliquot has become the benchmark for therapeutic PRC.<sup>4</sup> It should be noted that there is an optimal dose range of PRC. While application of the PRP enhances mesenchymal stem cell migration and proliferation, overexposure of cells to PRC yields many cells but limited differentiation of those cells into appropriate cell lines.<sup>12</sup> Opponents of the use of PRP cite the inability to control differentiation as a reason not to use this material in healing tissue.

In terms of processing, platelet collection should commence before surgery because activity at the surgical site will initiate clotting, thereby reducing the systemic platelet concentration. It is felt by some that even the initiation of an inhalation anesthetic agent will initiate the activation of platelets. Until the recent development of cell-sensitive filtration systems, centrifugation was the primary basis of producing a platelet-rich fraction. One of the disadvantages of centrifugation is that it can lead to fragmentation and lysis of the platelets, which triggers early release of growth factors and cytokines compromising bioactivity.<sup>13</sup> Fragmentation and early activation have also been shown with previous filtration-based systems that were either too vigorous or used pediatric dialysis filters in combination with cell saver, resulting in significant platelet degranulation and decreased efficacy.<sup>11</sup> Another disadvantage of centrifugation is that it requires the presence of additional capital equipment in the operating room. In some instances, the nursing or other ancillary staff may be required to transport the patient's blood out of the operating room for centrifugation.

The regenerative potential of PRP depends on the amount of growth factors and cytokines released when the platelets are activated. Growth factor and cytokine concentration is dependent on the concentration of these proteins in the platelets, the processing technique, which influences platelet concentration and hence protein concentration, and the completeness of platelet activation before measurement.<sup>14,15</sup> Before assessment of growth factor and cytokine concentration, these proteins must be released from platelets. Platelet activation and growth factor release can be accomplished by the addition of calcium or thrombin to the platelet concentrate. Studies have typically shown a 3- to 4-fold increase in growth factor concentrations in PRP as compared to nonconcentrated autologous blood.<sup>16</sup> Large variations exist between commercially available systems; therefore, each system needs to be tested individually for growth factor and cytokine expression. Once prepared, PRP is stable for 8 hours in the anticoagulated state. Before application, the concentrate should be activated by the addition of either 1000 units of topical thrombin or 10% calcium chloride. Exogenous thrombin and/or calcium can be added to PRP for immediate release of growth factors. Alternatively, endogenous release of these mediators by local tissue at the site of delivery can result in a slower release of growth factors and chemical mediators. Multiple delivery options exist, including single and dual syringes as well as direct transfer of the clot to the region of interest.<sup>1</sup>

PRC without centrifugation can be prepared using commercially available assay devices. Unlike previous filtration

devices (eg, AGF, Interpore Cross, Irvine, CA), platelet extraction and concentration are relatively atraumatic when using a cell-sensitive filtration device. Priming solution is injected into the device and mixed with 60 mL of anticoagulated blood from the patient. The blood and priming solution then flows through the filter. Approximately 7 mm of PRC is recovered by back-flushing the filter with a sterile syringe. In an unpublished study comparing a cell-sensitive filtration system (Caption; Smith & Nephew, Memphis, TN) to the centrifugation of blood, the processing time to obtain the filtrate was 40% faster and resulted in a similar concentration factor of platelets.

## Safety

Because it is an autogenous preparation, PRC is inherently safe and therefore free from concerns over transmissible diseases such as HIV, hepatitis, West Nile fever, and Creutzfeldt-Jakob disease. PRC, therefore, is well accepted by patients. There are some concerns about the use of bovine thrombin as the clotting initiator owing to the potential development of antibodies against the bovine thrombin. However, the antibodies developed against bovine factor Va, which has been essentially eliminated in the manufacturing and processing of bovine thrombin since 1997.<sup>17</sup> PRC use is contraindicated in patients who have preexisting coagulation defects (thrombocytopenia, hypofibrinogenemia, or on anti-coagulant therapy) or potentially have a hypersensitivity to bovine products.<sup>18</sup>

## Clinical Applications

The clinical use of PRP has been reported for a wide variety of applications, most predominately for the problematic wound, maxillofacial applications, and spine.<sup>10</sup> Collectively, these studies provide support for the clinical use of PRP. However, many reports are anecdotal, and few Level 1 studies with control group comparison are available to definitively determine the role of PRP. There is little consensus regarding the production and characterization of PRP, which can impede the establishment of standards that are necessary to integrate the enormous amount of literature from various studies on the subject.<sup>11,19</sup>

One of the first clinical applications of PRC was in the oral and maxillofacial surgical literature, where autologous fibrin adhesive was added to cancellous bone during mandibular continuity reconstruction. The study, published in 1994, revealed earlier radiographic bone consolidation (4 weeks versus 8 weeks), which was attributed to enhanced osteoconduction given to the osteocompetent cells in the graft by the fibrin network developed by the concentrated platelet formation.<sup>20</sup>

Another application of PRP is in the realm of hemostatic control. Hemostasis is important for any surgical procedure. One method uses autologous PRP sprayed on the wound site. A partial thickness skin wound model has shown PRP to reduce bleeding by 70 percent at 5 minutes when compared to placebo controls. Incidentally, platelet poor plasma has shown only a 10% reduction in bleeding in that same model.<sup>21</sup>

When the PRC is activated, proponents suggest that benefits include bone and soft-tissue restoration, wound healing, and a decrease in postoperative infection, pain, and

blood loss.<sup>10</sup> There have been numerous publications on the use of PRC for clinical applications in periodontal and oral surgery with sinus lifts, maxillofacial surgery with autogenous mandibular bone grafts, plastic surgery with face lifts and cosmetic dermal fat grafts, spinal fusion by enhancing graft formation in mixing with autogenous bone graft, heart bypass surgery, and the treatment of chronic skin and soft-tissue ulcers.<sup>10,20,22–25</sup> The quantity of the PRP used and the methods of application are procedure-specific. Furthermore, the research results are predominantly case studies or limited case series. It is unclear whether studies suggesting that there is no benefit from PRP are secondary to poor quality PRP produced by inadequate methods.<sup>4</sup>

### Orthopaedic Applications

PRC has significant potential in the treatment of pathologic conditions of cartilage, tendon, ligament, and bone. *In vitro*, there is a dose-response relationship between platelet concentration and the proliferation of human adult mesenchymal stem cells, proliferation of fibroblasts, and the production of type I collagen.<sup>26</sup> However, before widespread use of this technology can take place, further basic science and clinical research is needed to define the treatable musculoskeletal conditions, methods of administration, and ideal patient population. There are a number of orthopaedic studies that have been performed or are currently underway using PRC.

#### Tendon

A recent review of common growth factors suggested that PRP may be useful for tendon and ligament healing *in vivo*.<sup>27</sup> TGF- $\beta$  significantly increases type I collagen production in tendon sheath fibroblasts.<sup>28</sup> Previous animal studies have shown platelet-rich plasma to enhance Achilles tendon stiffness and force to failure in a rat model. Randomized prospective trials are underway in Europe to evaluate autologous PRP in Achilles tendon repairs.

In the first *in vivo* human investigation of autologous PRC as a treatment for chronic severe elbow tendinosis in patients who had failed nonoperative treatment, the data suggest buffered platelet-rich plasma may be an alternative to surgery in patients with this disorder. In Mishra's series, the fifteen PRP-treated patients with recalcitrant lateral epicondylitis demonstrated significant improvement with a single injection that was sustained over time with no reported complications.<sup>29</sup> Six-month data comparing PRC to cortisone injections in a prospective study show significant improvement with the autologous blood product in chronic tennis elbow, consistent with Mishra's previously reported findings.<sup>29</sup>

#### Bone

PRC has also been shown to be osteopromotive, rather than osteoinductive.<sup>30</sup> There are few basic science or clinical studies examining the role of PRC in bone healing after orthopaedic trauma.<sup>12,25,31</sup> Treatment of human mesenchymal stem cells in an osteoconductive environment with clotted PRC can enhance bone formation by modulating cellular pathways. Currently, it is common to combine the platelet-rich material with autograft, allograft, demineralized bone matrix, or other graft material. In fact, PDGF release from platelet rich concentrate was

markedly reduced in the presence of demineralized bone matrix. When PRP was applied in conjunction with autogenous bone graft, the rate of bone formation doubled and bone density increased by 25% when compared to controls.<sup>32</sup>

However, there have been some data presented stating that PRP has limited or even negative efficacy in certain delivery vehicles.<sup>30,33</sup> PDGF was shown to inhibit intramuscular osteoinduction and chondrogenesis by demineralized bone matrix in immunocompromised mice. PRP also reduced the osteoinductivity of active demineralized bone matrix. In a similar model, PRP decreased the osteoinductivity of demineralized bone matrix, and the activities of both demineralized bone matrix and PRP were donor-dependent. Yet, more recent data have shown the improved efficacy of PRC and bone graft materials on human bone marrow stromal cell activity, with bone formation being significantly modified by adding the agents in combination.

While mesenchymal stem cell proliferation can be upregulated by human serum, PRC derived from serum induces a more rapid response. Furthermore, PRC induces a rapid increase in mRNA production for BMP-2 and RUNX2, 2 growth factors involved in osteogenesis (unpublished data). Recent work by Sipe et al has even gone so far as to identify both BMP-2 and BMP-4 within platelet lysate, suggesting the possibility that this might contribute to the role of platelets in bone formation and repair.<sup>34</sup>

In summary, the initiation of bone regeneration starts with the release of PDGF and TGF- $\beta$  from the degranulation of platelets in the PRC. PDGF stimulates mitogenesis of marrow cells and endosteal osteoblasts. PDGF also initiates angiogenesis by inducing capillary budding into the surgical site through endothelial cell mitosis. TGF- $\beta$  activates fibroblasts, induces pre-osteoblasts to divide and increase in number, and triggers differentiation of pre-osteoblasts to mature osteoblasts. TGF- $\beta$  also induces osteoblasts to lay down bone matrix. Fibroblasts deposit a collagen matrix to support capillary ingrowth, which are visualized by day 3. By day 14, capillaries permeate the regenerative bone site. As this cellular activity is going on, growth factors are relied on to rapidly increase the numbers of these cells and promote their activity during time of injury and/or surgery. As the platelets come to the end of their life cycle, the growth factors have already activated chemotaxis, and macrophages have been triggered to replace the platelets as the primary source of factors. Macrophage-derived growth and angiogenic factors become the primary cellular drivers of bone healing. Marrow stem cells secrete TGF- $\beta$  to self-stimulate bone formation. Once the site is revascularized (after about 4 weeks), it is now self-sustaining. Maturation of the bone now comes from bone morphogenic protein produced by the bone matrix; as the matrix is formed and mineralized, BMP is laid down within the matrix. Bone morphogenic protein is released by osteoclastic resorption of normal bone remodeling, acting on stem cells to increase and differentiate into osteoblasts. This process of bone healing can potentially be accelerated by initiating the cascade of events early in the cycle with the use of PRC.<sup>4,32</sup> In fact, previous studies have suggested that PRP mediates only the early aspects of the bone repair process through an osteopromotive mechanism.<sup>12</sup>

## Nonunions

In one series, researchers were able to measure levels of PDGF and TGF- $\beta$  in the fracture hematoma of 24 patients who had fresh fractures of the foot and ankle; however, these investigators were unable to detect these same proteins in the nonunion tissue of 7 patients presenting with similar fractures. They prepared autologous PRP and were able to measure high concentrations in the concentrate of the missing growth factors in the nonunion sites. After application of the PRC to the nonunions during revision surgery, radiographic union was observed by an average of 8.5 weeks. Such studies, although not randomized and prospective, provide evidence of the utility of platelet therapy applied to nonunion patients or patients at high-risk for not healing their fractures.<sup>19</sup>

It should be noted that the use of PRP can help to augment fusions, but it does not eliminate the need for meticulous technique or the use of structural graft when required. The surgeon cannot expect PRC to promote adequate bone growth in an area of bone defect or void, but rather to augment the healing of 2 well-opposed adjacent surfaces, comprised mainly of cancellous or corticocancellous bone; currently, there is no evidence showing that pure cortical bone alone benefits from PRP.<sup>35</sup>

## Total Joint Arthroplasty

Biological materials used to assist in hemostasis after total knee arthroplasty have been the subject of much recent research. In a retrospective review of 98 total knee arthroplasty patients (61 of whom had PRC applied intraoperatively to exposed tissues, synovium, and the lining of the wound at closure), the patients receiving PRC during surgery had less postoperative blood loss, less oral and intravenous narcotic requirement, greater range of motion at discharge, and a shorter hospital stay than their counterparts who did not have the PRP applied to their wound. Further prospective trials with a placebo control arm and comparable treatment modalities are necessary, but this highly subjective observational study indicates that the application of platelet rich concentrate may lead to improved outcomes after total knee arthroplasty. Potentially, PRP applied directly to the operative site after knee replacement seals the tissues and delivers platelets directly to the wound.<sup>36</sup>

Another potential application in trauma or total joint arthroplasty involves use of PRP at the interface between the implant and the bone. With the decline in the use of cement, and the corresponding increased use of press-fit implants, PRP may promote earlier and more complete osteointegration of implants into host bone. Siebrecht and colleagues demonstrated that PRP, prepared from human blood using a commercially available platelet concentrate system, significantly increased bone and total tissue ingrowth distance compared with untreated controls in an athymic rat bone chamber model. The investigators theorized that porous hydroxyapatite lacks the cells and growth factors present in bone graft, but that PRC would potentially diminish this disadvantage.<sup>37</sup> This technology and technique is currently being applied in oral surgery, where implants are placed into extraction sites augmented with PRC.<sup>25</sup>

## Diabetic Fractures

The association between diabetes mellitus and impaired osseous healing has been documented in clinical and

experimental settings. Diabetes impairs the fracture healing process beginning with a reduction in early cellular proliferation, continuing with a delay in chondrogenesis, and ending with a decrease in the biomechanical properties of the fracture callus. Several clinical series have noted that the healing time for diabetic patients is approximately twice as long as that of nondiabetic patients.<sup>38</sup> In addition, diabetic patients undergoing elective arthrodesis had a significantly increased incidence of delayed union, nonunion, and pseudoarthrosis.<sup>39</sup>

In a diabetic fracture model study, a significant reduction in PDGF, TGF- $\beta$ 1, insulin-like growth factor, and VEGF expression was demonstrated in the diabetic fracture callus compared to the nondiabetic fracture callus.<sup>12</sup> The application of PRC restored early cell proliferation during healing to levels comparable to nondiabetic controls. Biomechanical testing revealed improved fracture healing in platelet-rich treated diabetic fractures compared to those in nontreated diabetic controls.<sup>12</sup> The percutaneous injection of PRC normalized early diabetic fracture callus, but the biomechanical properties were only partially restored in late diabetic fracture callus. These data are consistent with that seen clinically in diabetic patients having improved healing and decreased complications after ankle fusion when treated with a PRC.<sup>18</sup>

## Wound Healing

The application of autologous PRP can enhance wound healing, as has been demonstrated in controlled animal studies for both osseous and soft tissues.<sup>25,31</sup> Numerous clinical trials have reported favorable results in wounds treated with platelet-rich concentrate. One series observed 17 of 21 chronic lower extremity wounds reepithelize during a 9-week course of twice-daily wound treatment with platelet concentrate on a collagen base. In contrast, only 2 of 13 similar wounds treated with placebo healed. After crossover of the placebo group, the remaining 11 nonhealed wounds achieved epithelization on an average of 7 weeks.<sup>22</sup> A similar protocol in 171 patients with 355 wounds reported a 78% rate of limb salvage in patients for whom amputation was initially recommended.<sup>40</sup>

The use of PRC on split thickness skin graft donor sites is an emerging application in wound care. The concentrated platelet clot is placed on the donor site surface and retained by an occlusive dressing. When added as a clot, PRC induces early cellular proliferation when compared to serum without leading to cell overgrowth or the inhibition of cellular differentiation. Early results have shown that when the dressing is removed at 7 days, the donor site has the appearance of a 21-day placebo control. The site treated with platelet concentrate also shows nearly complete epithelialization. Revascularization of the donor site is enhanced by the angiogenic activity of PDGF and TGF- $\beta$ . Furthermore, the fibrin acts as a scaffold for epithelial migration. The rapid development of granulation tissue and epithelialization leads to a less prolonged crusting phase, less pain, and earlier return to normal activity.

## CONCLUSION

The enhancement of healing by the placement of supraphysiologic concentration of autologous platelets at the site of tissue injury or surgery is supported by basic science

studies. Research has revealed that the role of platelets is much more involved than simply “plug” formation; they are responsible for actively extruding growth factors, which initiate soft tissue healing, bone formation, and stem cell recruitment. These growth factors and cytokines are proteins stored in the alpha granules of the platelets, which are expressed with trauma or surgery.

There exist only a small number of controlled, clinical studies that provide evidence that the use of autologous PRC does accelerate soft-tissue and osseous healing in certain applications. However, PRC has been shown to enhance human mesenchymal stem cell proliferation in both in vitro and in vivo studies. The future of PRC in orthopaedic surgery remains largely anecdotal at this time. Improved preparation and salvage techniques, such as the use of a filtration device, as opposed to a centrifuge, along with controlled clinical studies will improve the application and potential efficacy of PRC in optimizing patient care with respect to bone and soft tissue healing. Proper clinical use and a well-developed understanding of the role that platelet-rich fractions can play in various orthopaedic clinical applications is key to achieving desired outcomes.

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