

## Platelet Rich Plasma: Myth or Reality?

Platelet rich plasma (PRP), also termed autologous platelet gel, plasma rich in growth factors (PRGF), platelet concentrate (PC), is essentially an increased concentration of autologous platelets suspended in a small amount of plasma after centrifugation. Basically, patient's blood is collected and centrifuged at varying speeds until it separates into 3 layers: platelet poor plasma (PPP), PRP, and red blood cells. Usually 2 spins are used. The first spin ("Hard spin") separates the platelet poor plasma (PPP) from the red fraction and platelet rich plasma (PRP). The second spin ("Soft spin") separates the red fraction from the PRP. The material with the highest specific gravity (PRP) will be deposited at the bottom of the tube. Immediately prior to application, a platelet activator/agonist (topical bovine thrombin and 10% calcium chloride) is added to activate the clotting cascade, producing a platelet gel. The whole process takes approximately 12 minutes and produces a platelet concentration of 3-5x that of native plasma.<sup>1,2</sup>

Platelets play a fundamental role in hemostasis and are a natural source of growth factors. Growth factors, stored within platelet  $\alpha$ -granules, include platelet derived growth factor (PDGF), insulin like growth factor (IGF), vascular endothelial growth factor (VEGF), platelet derived angiogenic factor (PDAF), and transforming growth factor beta (TGF- $\beta$ ). The release of these growth factors is triggered by the activation of platelets that can be initiated by a variety of substances or stimuli such as thrombin, calcium chloride, or collagen. Growth factors are involved in key stages of wound healing and regenerative processes including chemotaxis, proliferation, differentiation, and angiogenesis.<sup>3</sup> According to the definition of PRP, it may be assumed that these growth factors are present at increased concentrations in PRP. In addition to growth factors (GFs), platelets release numerous other substances (e.g., fibronectin, vitronectin, sphingosine 1-phosphate, etc...) that are important in wound healing. An advantage of PRP over the use of single recombinant human growth factor delivery is the release of multiple growth factors and differentiation factors upon platelet activation.<sup>4</sup> Recently, the morphologic and molecular configuration of PRP was reported, it showed PRP is a fibrin framework over platelets that has the potential to support regenerative matrix.<sup>5,6</sup>

The rationale for using PRP in soft and hard augmentation are to accelerate vascularization of the graft, improve soft tissue healing, reduce post operative morbidity, and enhance bone regeneration.<sup>7</sup> Advantages of using an autologous PRP include no risk of cross reactivity, immune reaction or disease transmission.<sup>8</sup> In addition, the use of PRP improves handling of particulate

graft materials and easier packing into a grafting site, thus facilitating space maintenance and potential bone regeneration.<sup>9,10</sup>

Since PRP contains several GFs (e.g., PDGF, VEGF, etc...) that are capable to stimulate angiogenesis and increase fibroblast cell differentiation, using PRP to promote soft tissue healing has been proposed.<sup>2</sup> Research showed that PRP and analogous products improve graft adhesion and minimizes micro-movement, providing the most advantageous environment for graft acceptance.<sup>11,12</sup> It has been also proposed that PRP accelerates wound maturity and epithelialization, hence decreased scar formation. PDGF and epidermal growth factor (EGF) are the main growth factors involved in fibroblast migration, proliferation, and collagen synthesis. Increased concentrations of these growth factors are likely the reason for the accelerated soft tissue wound healing, which is suggested to be at least 2-3 times faster than that of normal.<sup>13</sup>

For the hard tissue, growth factors released from PRP are likely to effect local vital cells such as osteoblasts. The addition of PRP to stromal cells has demonstrated angiogenic and osteogenic properties in animal models.<sup>14</sup> The use of PRP to enhance bone regeneration has been documented in periodontal defects,<sup>15-17</sup> extraction sockets,<sup>18</sup> during implant placement,<sup>19,20</sup> and in guided bone regeneration procedures around implants, including sinus augmentation.<sup>21</sup>

One of the major drawbacks of bone augmentation is the extended healing time required. Hence, one of the major reasons proposed for the use of PRP is a reduced healing time. A shortened graft healing time (50%) has been demonstrated in sinus augmentation.<sup>22</sup> Accelerated bone regeneration has also been demonstrated in periodontal defects distal to second molars when PRP is added at the time of extraction of impacted third molars.<sup>18</sup> Unfortunately these results cannot be used to expound the beneficial effects of PRP, as biopsies were not taken from any of the control sites.

In animal studies, when cancellous bone from the iliac crest was used as graft material for sinus lifts with or without PRP, biopsies showed both PRP and control groups achieved similar results with no statistically significant difference between the two.<sup>9,23</sup> It is also true when DFDBA was used as a graft material, PRP did not enhance its ability to form bone.<sup>24</sup> Similar findings were also reported when PRP was added to the xenograft (e.g., Bovine HA).<sup>25</sup> These results are consistent with those above that failed to demonstrate enhanced bone regeneration when PRP is combined with non-living graft materials.

In humans, early controlled studies demonstrated

enhanced healing and bone regeneration with PRP application.<sup>1,7</sup> A short-term increase (e.g., improved at 2 weeks but not at 12 weeks) of bone regeneration in sinus augmentation was observed when compared the  $\beta$ -TCP with or without PRP.<sup>26</sup> Similar results have been demonstrated in minipigs,<sup>25</sup> and adult domestic pigs.<sup>27</sup>

When FDBA was used with PRP for subantral sinus augmentation, results showed the application of PRP will only result in accelerated new bone formation if target cells such as osteoblasts and osteocytes are present.<sup>28</sup> Also, study has reported that, when combined with anorganic bovine bone grafts, the addition of PRP had no effect on defect mineralization at any time point.<sup>26</sup> This may be one of the reasons that several studies utilizing non vital grafting material have failed to show any beneficial effect.<sup>25,29,30</sup>

Numerous reports have been published regarding the favorable effects of PRP on wound healing after bone augmentation. However, many are case reports lacking controls.<sup>31-34</sup> Hence, a standardized research protocol should be used for future studies. These include control of the quantification of platelet yield in both whole blood and PRP, use of commercial assays to quantify growth factor concentration, utilization of contralateral control groups, and proper histomorphometric analysis of specimens. The correlation between GF levels and histomorphometric result is another area that needs to be examined. One of theories behind why PRP does not work may be the concentration of PRP from the machine is too low for the GFs to show the clinical effect. It would be interested to know if the concentration is increased to 10 times (versus 3-4 times) over the plasma, what kind of clinical results illustrated.

Safety of PRP remains to be an issue since increases GF in a local area may be a cancer promoting effects. Other concern is the use of bovine thrombin in activating the PRP release to cause immunogenic reaction.<sup>35</sup> So far most of published data showed it is safe to use the product, however, future study in this area is certainly needed.

Currently there is a paucity of critical scientific data regarding the beneficial effects of platelet rich plasma in clinical procedures. There have been animal and human studies both purporting and refuting its adjunctive positive effect. In theory, PRP has many beneficial effects such as autologous supply of growth factors and improved wound healing. In addition it is relatively inexpensive and readily available. However, from the current available literature it is clear that there is great variability in study design, clinical and radiographic parameters that were measured, and clinical outcome. Many studies claiming a positive beneficial effect suffer from a poor study design. Many have no controls or a limited sample size. In general, conclusions advocating for an adjunctive effect are not supported by the study design. Therefore, the use of this material cannot be supported at present, and further controlled, prospective clinical trials are urgently needed.

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