INTRODUCTION:

While the healing effects have not been consistent in multiple models and studies, Autologous Platelet-Rich Plasma (PRP) has been shown in some studies to improve soft tissue and bone healing.\(^1\)\(^-\)\(^3\) It is presumed that any positive effect is achieved by delivering a concentrated amount of growth factors such as transforming growth factor-β1 (TGF-β1); platelet-derived growth factor (PDGF) – AA, AB, BB; and vascular endothelial growth factor (VEGF), contained in platelet α-granules. These growth factors, among others induce chemotaxis to and mitogenesis of macrophages, fibroblasts, and osteoprogenitor cells within the wound and fracture.\(^1\)\(^-\)\(^8\)

In this study our aim was to compare platelet concentration variability in whole blood (WB) and manually prepared PRP in the female New Zealand White rabbit model. In addition, we set out to examine the following:

1. Correlation between PRP platelet concentration and arterial whole blood platelet concentration
2. Correlation between arterial whole blood platelet concentration and rabbit weight
3. Correlation between arterial whole blood platelet concentration and rabbit age

METHODS:

This study protocol was approved by our institutional IACUC. 56, 3-5 kg female New Zealand White Rabbits ages 6 – 33 months were identified for whole blood platelet analysis and potential PRP treatment.\(^4\) Immediately following general anesthesia, a 17-gauge needle was inserted into the central auricular artery and 10 ml autologous arterial blood was drawn into a sterile citrate tube. PRP was manually prepared using two centrifugations (300g for 10 minutes to separate out red blood cells and 5,000g for 5 minutes to separate out platelet poor plasma).\(^5\)\(^-\)\(^9\) Whole blood and PRP platelet concentration was quantified by standard veterinary laboratory analysis.

Statistical analysis was performed using SAS statistical software version 9.2 (SAS Institute Inc., Cary, NC, USA, 2008). Four samples were excluded from the study (two due to the formation of clots during PRP preparation and two data points were identified as statistical outliers).

RESULTS:

Table 1: Mean Values, Ranges, and Enrichment of Platelets (PLT)\(^9\)

<table>
<thead>
<tr>
<th></th>
<th>PLT concentration (x 10^3/µL)*</th>
<th>PLT range (x 10^3/µL)</th>
<th>Enrichment range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arterial WB</td>
<td>247 ± 83</td>
<td>121 - 452</td>
<td>-</td>
</tr>
<tr>
<td>PRP</td>
<td>1188 ± 421</td>
<td>497 - 2409</td>
<td>2.89 - 8.80</td>
</tr>
</tbody>
</table>

\(^*n = 52, *mean ± SD\)

Table 1 represents our observed mean values ± standard deviation and ranges of platelet concentration in rabbit arterial whole blood and PRP, and enrichment concentration range of PRP (PRP/WB).

There was a positive correlation between the arterial whole blood platelet concentration and prepared PRP platelet concentration (R=0.769, P < 0.05) shown in Figure 1.

There was a weak positive correlation between the age of the rabbit and arterial whole blood platelet concentration (R=0.397, P < 0.05) shown in Figure 2.

There was no statistical correlation between arterial whole blood platelet concentration and the weight of the rabbit at the time of blood draw (P < 0.05).

DISCUSSION:

We observed an arterial whole blood platelet concentration range of 121 – 452 x 10^3 platelets/µL. Current literature suggests New Zealand White rabbit venous whole blood platelet concentration ranges from 250 - 750 x 10^3 platelets/µL and human whole blood platelet concentration ranges from 150-450 x 10^3 platelets/µL.\(^7\)\(^-\)\(^14\)

As expected, there was a strong positive correlation between PRP platelet concentration and arterial whole blood platelet concentration (R=0.769, P < 0.05) as shown in Figure 1. Given the variation in platelet number in whole blood, for the same platelet concentration percentage, the total number of platelets in the PRP is expected to vary.

A weak positive correlation between whole blood platelet concentration and rabbit age was observed as shown in Figure 2. Future studies should investigate this observation in greater detail.\(^12\)\(^13\)

It is outside the scope of this study to examine arterial whole blood platelet concentration drawn from the central auricular artery compared to venous whole blood platelet concentration drawn from the marginal ear vein, though data suggests that there may be a significant difference in platelet concentration between the two.\(^1\)\(^-\)\(^4\)\(^-\)\(^9\)\(^-\)\(^10\) Future models and studies should investigate this issue.

SIGNIFICANCE:

Understanding platelet variability in whole blood and PRP will allow clinicians and researchers to develop standardized PRP protocol and better understand the application of PRP in bone healing.

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