A Prospective Comparison of the Platelet Sequestration Ability of Three Autotransfusion Devices

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Abstract: Although current autotransfusion devices have platelet sequestration capabilities, each has a unique technology to achieve the final platelet product. The purpose of this study was to assess the quality and quantity of platelets sequestered by three different autotransfusion devices. The three commercially available autotransfusion devices evaluated were Fresenius C.A.T.S (closed spiral chamber), Cobe BRAT 2 (Baylor bowl), and Haemonetic Cell Saver 5 (Latham bowl). Platelet sequestration was performed in the automatic mode following the manufacturer’s recommended sequestration protocols. The total number of platelets sequestered, percent recovery, and percent platelet function were assessed. Each device behaved similarly. There was a 2- to 3-fold increase in platelet count compared with baseline with only approximately 50–60% recovery, whereas there was a 10% decrease in platelet function after processing compared with baseline. No statistical difference was noted in platelet function between the respective machines. However, there was a significant loss of platelet function observed with the actual process regardless of autotransfusion device used. **Keywords:** platelet sequestration, auto transfusion devices, platelet function.

The benefits of limiting a patient’s exposure to homologous blood products have been documented (1–4). Thus, blood conservation techniques are becoming increasingly popular in today’s clinical setting. In the operating room, one method of blood conservation has been autologous blood component therapy. Autotransfusion devices can efficiently separate a unit of the patient’s own blood into red blood cells (RBC), platelet-poor plasma (PPP), and platelet-rich plasma (PRP). These individual products can then be administered back to the patient postoperatively or as required. This technique has been shown to be very beneficial in high-risk cardiac surgery patients through the preservation of the patient’s own platelets resulting in the reduction of homologous platelet transfusions (5). Autotransfusion devices are becoming a very common piece of equipment in the operating room. Even though these devices were originally designed for cell salvaging, most machines now have platelet sequestration capabilities available for the production of PRP. All use centrifugation to separate whole blood into its components, however each uses different technology to accomplish this.

Historically, hospitals have used a platelet count to assess the quality of the PRP product. However, platelet count does not verify the quality of platelets produced or how well they are functioning. Sonoclot Coagulation & Platelet Function Analyzer (Sienco Inc., Morrison, CO) and the thromboelastograph (Haemoscope Corporation, Niles, IL) measure the hemostatic changes in clot formation (6,7). However, Helena Laboratories (Beaumont, TX) has developed and introduced a two-step platelet function point of care assay called Platelet Works (8) that gives the user a quantitative number applied to the functioning platelets. Through the activation of platelets by an agonist and comparing that with an initial platelet count, the percentage of a person’s functioning platelets can be determined at the bedside. The purpose of the study was to investigate whether there was an observable deviation in the quality of PRP produced by three different autotransfusion devices. As well, to assess the affect the process itself and storage has on platelet function.

**MATERIALS AND METHODS**

The hospital internal review committee approved the study protocol. Platelet sequestration was performed in the operating room on adult patients undergoing cardiac surgery. Patients requiring cardiac surgery were prospectively assigned to one of three autotransfusion devices.
Patients were excluded from the study if they had a weight less than 50 kg, platelet counts < 150 × 10$^3$/mm$^3$, a hemoglobin < 120 g/dL, used acetylsalicylic acid within 7 days, used Plavix (clopidogrel bisulfate) within 30 days, had known platelet dysfunction, had critical aortic stenosis with a valve area <0.7 cm$^2$, New York Heart Association class 3-4 left ventricular function, or left main coronary artery disease >75%.

Before skin incision, blood was drawn from a 20-gauge arterial line catheter for initial platelet assessment (sample 1). Simultaneously the patient was passively phlebotomized through a high-flow stopcock connected to a 9 FR central line. Citrate phosphate dextrose adenine (CPDA–1) solution was used (63 mL) as an anticoagulant in a blood-pack unit. Each CPDA–1 bag was placed on a Sebra automatic rocker weigh scale (Model 1020; Tucson, AZ) to ensure a precise volume of 450 mL of blood was removed from the patient.

The autologous blood was then immediately processed into PRP, PPP, and RBC using one of three autotransfusion devices. The auto transfusion devices evaluated were: (1) Cobe Brat 2 - Baylor Rapid Autologous Transfusion System (Sorin Biomedica, Arvada, CO); (2) Fresenius C.A.T.S - Continuous AutoTransfusion System (Fresenius HemoCare GmbH, Bad Homburg, Germany); and (3) Haemonetics Cell Saver 5 - Autologous Blood Recovery System (Haemonetics Corporation, Braintree, MA).

Platelet sequestration was executed following the manufacturer’s programmed recommendations (Table 1). The same device was used throughout the study and only properly trained personnel performed the sequestration procedure.

PRP was stored at room temperature on an automatic rocker. A second sample was drawn from the PRP bag using a luered tip syringe upon completion of platelet sequestration. A third sample was drawn from the same PRP bag approximately 2 hours after processing.

Samples were assessed for quantity and quality of platelets using Platelet Works Point-of-Care Analyzer. The two-step assessment involves the use of a baseline as well as an agonist test tube. Ethylene diamine tetraacetic acid (EDTA) was used in the baseline test tube whereas adenosine diphosphate (ADP) was used as the agonist. The ADP activates any functioning platelets, causing their aggregation, thus removing them from the agonist platelet count. Therefore, the platelets that are counted in the agonist tube are the nonfunction platelets. To minimize platelet destruction the transfer of each sample was achieved by removing the cap off the vacuum-sealed tubes and the correct volume was ejected from the syringe to slide down the side of the tube. Each sample was treated in the same manner.

**Sample Analysis**
All samples were analyzed for platelet function and recovery using the following equations:

**Platelet Function:**

\[
\text{Platelet Function} = \frac{\text{absolute platelet count} - \text{agonist platelet count}}{\text{absolute platelet count}} \times 100\%
\]

Where the absolute platelet count is the baseline platelet count in the EDTA nonagonist test tube and the agonist platelet count is the platelet count of the agonist test tube. These are platelets that do not react to a stimulus (ADP); therefore, they are considered nonfunctioning.

**Percent Recovery:** Percent recovery is defined as the absolute number of platelets in the PRP as a percentage of the absolute number of platelets in the volume of blood processed to produce the PRP (absolute platelet count of PRP × volume of PRP)/(preop absolute platelet count × volume of blood processed for PRP procedure).

**Concentration Factor:** The ability of the autotransfusion device to concentrate platelets was determined using the following equation:

\[
\text{Absolute platelet count of PRP/absolute platelet count of the patient.}
\]

**Statistical Analysis**
Statistical analysis was done using a one-way analysis of variance. A two-tailed $p$ value of less than 0.05 was considered statistically significant.

**RESULTS**

**Demographics**
The number and type of procedures done for each device are summarized in Table 2.

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**Table 1. Manufacturer’s recommended platelet sequestration protocol.**

<table>
<thead>
<tr>
<th>Device</th>
<th>PPP Fill Rate/RPM</th>
<th>PRP Fill Rate/RPM</th>
<th>Extended PRP Collection</th>
<th>Type/Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cobe</td>
<td>50 mL/min/4400</td>
<td>25 mL/min/2400</td>
<td>Automatic</td>
<td>Baylor bowl/135 mL</td>
</tr>
<tr>
<td>Fresenius</td>
<td>75 mL/min/2000</td>
<td>100 mL/min/2000</td>
<td>15 mL</td>
<td>Spiral Chamber/300 mL</td>
</tr>
<tr>
<td>Haemonetics</td>
<td>60 mL/min/4750</td>
<td>60 mL/min/4750</td>
<td>30 mL</td>
<td>Latham turbo fin bowl/125 mL</td>
</tr>
</tbody>
</table>

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Platelet Count

As seen in Table 3, there was no statistical significant difference between the patient’s preoperative platelet counts (sample 1). Similarly, there was no difference between the initial PRP platelet counts (sample 2) of each machine group or the platelet count after 2 hours of storage.

Platelet Function

All patient’s had a similar preoperative platelet function (sample 1). The initial platelet function of the PRP sample (sample 2) showed similar results for the individual devices. However, there was a statistically significant drop in platelet function when comparing the preoperative platelet function and platelets functioning after sequestration. The 2-hour storage time did not have any affect on platelet function (Figure 1).

Concentration Factor

The Cobe, Fresenius, and Haemonetics machines were able to concentrate the original platelet count by 2.3 ± 0.6, 2.6 ± 1.1, 2.4 ± 0.6, respectively (Figure 2). There was no statistical significant difference seen between any of the groups.

Platelet Recovery

There was no statistical difference between autotransfusion devices with regards to absolute or effective platelet recovery (Figure 3). The absolute recovery of the Cobe was 57 ± 15%, followed by the Haemonetics with 56 ± 9%, whereas the Fresenius recovered 51 ± 23%.

DISCUSSION

Autotransfusion devices originally were designed to salvage red blood cells lost during surgical procedures. The platelet sequestration feature of an autotransfusion device was the industry’s attempt to remodel an existing device into a portable blood fractionation system. A multipurpose machine provides convenience, but the efficiency of the procedure may be compromised. As shown by this clinical trial, only 50% of drawn platelets were recovered by using an autotransfusion device to produce PRP. Fresenius and Cobe both document an average of 51% and 60% recovery rate, respectively (9,10). However, this does not take into account the 10–12% loss of platelet function caused by the process itself as identified in this study. This results in an effective recovery rate of less than 50%.

All of the manufacturers report an ability to concentrate platelets 3 times their original count. In this study the average platelet yield was approximately 2.5 times the original platelet count, and was highly inconsistent ranging from 1.4 to 3.6 times. One technical aspect of platelet sequestration using bowl technology is that the lower the hematocrit of the autologous whole blood the larger volume of blood that is required to complete a cycle. When processing a larger volume of blood a higher platelet yield is expected, however this was not observed in this study. We observed that a larger volume of blood was required to yield platelets when there was a low preoperative hematocrit regardless of the device being used. Even then this did not guarantee a concentration factor greater than 2.5.

Another observation in this study was that a 2-hour storage of the PRP produced by three autotransfusion devices did not affect platelet function. The Canadian Society for Transfusion Medicine and the AABB standards for storing platelets is a 5-day shelf life, incubated at 20–24°C with constant gentle motion (11,12). Although the temperature in the operating room varied from 18°C to 21°C, the other criteria were met with no significant effect on the quality of PRP produced.

One of the obvious limiting factors of this study was the small sample size. The difference between devices with only a small size of four to six patients has to be substantiated to see a significant difference. However, the sample size was enough to show that there is a significant decrease in platelet function attributed to the sequestration process.

In Conclusion, as shown by this study the efficacy of platelet preservation using autotransfusion devices is questionable. Indeed, several other studies have looked at

<table>
<thead>
<tr>
<th>Device</th>
<th>No. of Procedures</th>
<th>Type of Procedure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cobe: Brat 2</td>
<td>4</td>
<td>AVR (3), MVR, MVR, MVR + ACB, LVA + ACB, Redo ACB (3)</td>
</tr>
<tr>
<td>Fresenius: C.A.T.S.</td>
<td>4</td>
<td>MVR + ACB, Redo ACB</td>
</tr>
<tr>
<td>Haemonetics: Cell Saver 5</td>
<td>6</td>
<td>MVR + ACB (2), Redo ACB</td>
</tr>
</tbody>
</table>

Table 2. Number and type of procedure used with each device.

Table 3. Total platelet counts and platelet function of each sample interval.

<table>
<thead>
<tr>
<th></th>
<th>Platelet Count (×10⁴/mm³)</th>
<th>Platelet Function</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sample 1</td>
<td>Sample 2</td>
</tr>
<tr>
<td>Fresenius (n = 4)</td>
<td>237 ± 134</td>
<td>520 ± 86</td>
</tr>
<tr>
<td>BRAT 2 (n = 4)</td>
<td>187 ± 28</td>
<td>414 ± 84</td>
</tr>
<tr>
<td>Cell Saver 5 (n = 6)</td>
<td>168 ± 53</td>
<td>405 ± 175</td>
</tr>
</tbody>
</table>

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pre-operative plasmapheresis with subsequent reinfusion of PRP post-operatively as a blood conservation technique (13,14). Wong et al., found no benefit from PRP reinfusion post cardiac surgery (15). In isolated patients with excessive coagulopathy post cardiopulmonary bypass, Tobe et al., found a favorable effect with PRP reinfusion on coagulation and blood loss (14). Only Stammers et al. validated the use of this procedure in all cardiac surgery patients. However, the positive hematological benefits were short lived (5). To preserve platelet function, a more effective method may be to phlebotomize the patient for a unit of blood and re-transfuse the patient immediately post-operatively. This may result in a minimal loss of platelet function and a higher recovery.

REFERENCES