BACKGROUND: Six filters were tested for their ability to remove white cells from buffy coat-depleted red cell concentrates at various temperatures.

STUDY DESIGN AND METHODS: Cellselect FR, BPF4, and Sepacell filters were tested at both room temperature (RT) and 4°C. The Leucoflex filter was tested only at 4°C, while the Cellselect Optima Plus and Imugard filters were tested only at RT. Donor-dependent differences were excluded by pooling and subsequently dividing 9 red cell concentrates; 12 sets of experiments were performed.

RESULTS: With all filters, red cell concentrates containing <5 x 10^6 white cells per unit were obtained. The lowest numbers of residual white cells were achieved with the Leucoflex (at 4°C, 0.15 ± 0.11 x 10^6), the Sepacell (at 4°C, 0.23 ± 0.14 x 10^6), the Imugard (at RT, 0.24 ± 0.14 x 10^6), and the BPF4 (at 4°C, 0.25 ± 0.24 x 10^6; differences not significant). With the Cellselect FR, filtration at 4°C resulted in 0.86 ± 0.37 x 10^6 white cells per unit, a level not significantly different from that obtained with the BPF4 and Sepacell filters at RT (1.16 ± 0.43 x 10^6 and 0.80 ± 0.36 x 10^6 white cells, respectively). Filtration at RT with the Cellselect FR and Cellselect Optima Plus resulted in red cell concentrates with 1.79 ± 0.69 x 10^6 and 2.29 ± 0.69 x 10^6 white cells, respectively (p<0.01).

CONCLUSION: All filters conformed to the current standards for white cell reduction; the process was less efficient at RT than at 4°C. For routine application, the composition of the red cell concentrate, the temperature, and logistic preferences should be taken into account in the final choice of filter; before implementation, the chosen filter must be validated under routine conditions.

ABBREVIATIONS: BC = buffy coat; RBC(s) = red cell(s); RT = room temperature; WBC(s) = white cell(s).

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with 63 mL of CPD and in one satellite bag with 100 mL of saline-adrenaline-glucose-mannitol as additive for the RBCs (Biopack-Compoflex, NPBI, Emmer-Compascuum, the Netherlands). All whole-blood donations were cooled to 20 to 24°C under 1.4 butane-diol plates, as previously described.18

The following day, between 18 and 24 hours after blood collection, the whole blood was centrifuged for 10 minutes at 2960 × g in a centrifuge (J6MI, Beckman, Palo Alto, CA) at RT. Whole blood was then separated into plasma, BC, and BC-depleted RBC concentrates, using an automated blood component separation device (Compomat G4, NPBI).

### Pooling and dividing of the RBCs

To exclude donor-dependent differences, for each experiment, 9 BC-depleted RBC concentrates, with identical ABO and Rh blood groups, were pooled and subsequently divided into the original bags. A 3-L pool bag (NPBI) was connected with the appropriate tubing by the use of nine leads. Onto each lead, an RBC bag was welded by using a sterile connection device (SCD-201 A, Terumo, Tokyo, Japan). The contents of the RBC bags were transferred to the pool bag. The RBC pool was mixed thoroughly for at least 1 minute, and the contents were returned to the original RBC bags. Care was taken that the bags were filled uniformly, and the pool bag was mixed regularly to prevent sedimentation of the cells. After completion of this process, the bags were disconnected by heat-sealing, and the units were weighed.

A 150-mL transfer bag (Biopack-Compoflex, P4160, NPBI) was welded to each of the RBC bags. After thorough mixing, a sample of approximately 10 mL was obtained from each of the 9 units for in vitro analysis.

### Filtration of the BC-depleted RBC concentrates

All steps of the filtration protocol were performed according to the instructions of the manufacturers. The following filters were used: Leucoflex LCG1 (FEM 4007 A, lot no. 2908 97A23; MacoPharma, Tourcoing, France), Cellselect FR (B3118, lot no. 96K29; NPBI), Cellselect Optima Plus (B2145, lot no. 9507050707; NPBI), BPF4 (BPF4BBS, lot no. 96H23LO1; Pall, Newquay, UK), Sepacell (RZ-200Bl, lot nos. 259 TXF and 661 U2A; Asahi Medical Co., Tokyo, Japan); the whole chamber (50 μL) was counted. Absolute numbers of RBCs, WBCs, and platelets per unit were calculated from the volume and the cell counts.

The BPF4 and the Sepacell had to be filled by squeezing of the RBC bag. All other filters were filled spontaneously. All filtrations were performed by hanging the RBC bag approximately 1 m above the collection bag.

Air was expelled from the collection bag when the RBC bag was empty. Air was returned from the collection bag to the RBC bag through the air bypass, and it was used to remove the last blood from the filter. The air bypass possessed a one-way valve to prevent blood flow through the bypass. Filtration was terminated when no blood was visible on the entrance side of the filter (except with the Optima Plus filter, which did not have transparent housing: in it, filtration was terminated when no blood flow from the filter to the collection bag was visible).

After filtration, the weight of the filtered units was measured. A total of 12 sets of experiments was performed.

### WBC concentration in BC-depleted RBC concentrates after quick cooling and rewarming

To investigate whether the quick-cooling procedure had a deleterious effect on WBCs, 9 BC-depleted RBC concentrates were quickly cooled and rewarmed. With a syringe, 5-mL samples were taken before cooling, after the bags were placed under a cooled butane plate for 2 hours, and again after rewarmed for 16 hours. The samples were analyzed within 10 minutes after drawing.

### In vitro measurements

The volume of the RBCs was calculated by dividing the net weight of the content of the bag by the specific gravity of the units (1.070 g/mL at 60% hematocrit). Samples from the filtered RBCs were obtained by cutting the tubing after thorough mixing. Hematocrit values; RBC, WBC, and platelet counts; and hemoglobin content were determined with an analyzer (CA 570, Medonic, Bromma, Sweden). The volume of RBCs was calculated from the hematocrit and the total volume of the unit.

WBCs in the filtered RBCs were counted in a Nageotte hemocytometer, using a 1-in-5 dilution in a lysing solution (Leucoplate, Labo International, Maarssen, the Netherlands); the whole chamber (50 μL) was counted. Absolute numbers of RBCs, WBCs, and platelets per unit were calculated from the volume and the cell counts.

### Statistical analysis

All results were entered into a computer, and, by using a software program (Instat version 2.04, GraphPad Software, San Diego, CA) for statistical analysis, the data were compared. We used the repeated-measures ANOVA for paired samples (except when stated otherwise) and then used a Tukey-Kramer multiple-comparisons test to determine the difference between individual filters. Effective pairing of the samples was determined as a significant variation among means; p<0.05 was considered significant.
Although slightly higher numbers of residual WBCs were twice that with filtration at 4°C. The Cellselect FR produced obtained by filtration with the Sepacell at 4°C, the Imugard less efficient; the number of residual WBCs was at least cant. at RT, and the BPF4 at 4°C, the differences were not signifi-
cated RBCs with the lowest number of WBCs per unit. The Leucoflex produced AlI tested filters were able to reduce the WBC number to 109 per unit. The Leucoflex gave the best performance with all filtered RBCs containing <0.5 × 106 WBCs per unit.

As noted earlier, the number of platelets before filtration was already below our specifications for filtered RBCs, which should be taken into account in the interpretation of the following observation. Platelet removal was shown to be independent of filtration temperature (see Table 2); it averaged 25 percent for all filters tested, except for the BPF4, which removed only 10 percent of the platelets originally present (p<0.05).

### Volume and hemoglobin loss after filtration

During filtration, a volume of blood is lost in the housing of the filter. Because of the large housing of the Optima Plus, 10 out of 12 filtered RBC units had a total volume below the specifications (i.e., <225 mL). The Leucoflex produced 6 filtered RBC units with a low volume. All other units conform to the specifications.

### TABLE 1. Composition of BC-depleted RBC concentrates after pooling and subsequent division into 9 units, represented as mean ± SD (n = 12)

<table>
<thead>
<tr>
<th></th>
<th>Leucoflex (4°C)</th>
<th>Cellselect FR</th>
<th>Optima Plus BPF4</th>
<th>Sepacell</th>
<th>Imugard (20°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(A)</td>
<td>(B)</td>
<td>(C)</td>
<td>(D)</td>
<td>(E)</td>
</tr>
<tr>
<td>Volume (mL)</td>
<td>272 ± 9</td>
<td>273 ± 8</td>
<td>270 ± 7</td>
<td>271 ± 8</td>
<td>272 ± 7</td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>58.4 ± 1.0</td>
<td>58.5 ± 0.7</td>
<td>58.9 ± 0.8</td>
<td>58.8 ± 1.2</td>
<td>58.8 ± 0.9</td>
</tr>
<tr>
<td>RBC volume (mL)</td>
<td>159 ± 7</td>
<td>160 ± 6</td>
<td>160 ± 6</td>
<td>159 ± 7</td>
<td>160 ± 6</td>
</tr>
<tr>
<td>RBCs (×10¹²)</td>
<td>1.76 ± 0.07</td>
<td>1.77 ± 0.07</td>
<td>1.76 ± 0.06</td>
<td>1.77 ± 0.08</td>
<td>1.77 ± 0.07</td>
</tr>
<tr>
<td>Platelets (×10¹²)</td>
<td>6.4 ± 1.0</td>
<td>6.8 ± 1.7</td>
<td>7.1 ± 1.6</td>
<td>7.8 ± 2.0</td>
<td>6.6 ± 0.9</td>
</tr>
<tr>
<td>WBCs (×10¹²)</td>
<td>586 ± 150</td>
<td>606 ± 160</td>
<td>583 ± 140</td>
<td>596 ± 155</td>
<td>589 ± 152</td>
</tr>
<tr>
<td>Hemoglobin (g)</td>
<td>50.4 ± 2.6</td>
<td>50.6 ± 2.1</td>
<td>50.8 ± 2.0</td>
<td>50.4 ± 2.4</td>
<td>50.8 ± 2.3</td>
</tr>
</tbody>
</table>

* Letters A through I are assigned to a combination of filter and temperature to facilitate interpretation of results.

### TABLE 2. Residual WBCs and platelets in filtered RBCs, after filtration of BC-depleted RBC concentrates with comparable composition, as obtained by pooling and dividing

<table>
<thead>
<tr>
<th></th>
<th>Leucoflex</th>
<th>Cellselect FR</th>
<th>Optima Plus</th>
<th>BPF4</th>
<th>Sepacell</th>
<th>Imugard (20°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(4°C)</td>
<td>(20°C)</td>
<td>(4°C)</td>
<td>(20°C)</td>
<td>(4°C)</td>
<td>(20°C)</td>
</tr>
<tr>
<td>WBCs†</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± SD (×10⁶)</td>
<td>0.15 ± 0.11 1.79 ± 0.69 0.86 ± 0.37 2.29 ± 0.69 1.16 ± 0.43 0.25 ± 0.24</td>
<td>0.80 ± 0.36 0.23 ± 0.14 0.24 ± 0.14</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range (×10⁹)</td>
<td>&lt;0.02 ± 0.36 0.78 ± 2.96 0.32 ± 1.40 1.17 ± 3.48 0.52 ± 1.48 0.02 ± 0.86</td>
<td>0.24 ± 1.55 0.07 ± 0.53 0.11 ± 0.59</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean log₁₀ reduction</td>
<td>3.68 2.55 2.86 2.42 2.72 3.52</td>
<td>2.90 3.46</td>
<td>3.43</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of units with &lt;5 × 10⁶</td>
<td>12 12 12 12 12 12</td>
<td>12 12 12</td>
<td>12</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of units with &lt;1 × 10⁶</td>
<td>12 2 8 0 4 12</td>
<td>9 12</td>
<td>12</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of units with &lt;0.5 × 10⁶</td>
<td>12 0 3 0 0 10</td>
<td>3 11 11</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Platelets†</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± SD (×10⁹)</td>
<td>4.5 ± 0.7 5.4 ± 1.1 4.6 ± 1.1 4.4 ± 0.5 5.9 ± 1.7 5.8 ± 0.8</td>
<td>5.4 ± 0.7 5.0 ± 0.7 4.7 ± 1.4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Results reported as mean ± SD (n = 12).
† All differences p<0.001, except A vs. F, H, and I (NS); F vs. H and I (NS); H vs. I (NS); C vs. E and G (NS); and B vs. D (p<0.01).
‡ All differences NS, except A vs. E and F (p<0.05); C vs. F (p<0.05); D vs. F (p<0.05); and D vs. E (p<0.01).
Except for the Optima Plus, in which the hematocrit after filtration was lower than that before filtration (p = 0.01, paired t test; see Table 3), RBCs were not specifically retained by the filters. Therefore, volume loss strongly correlated with the number of RBCs lost during filtration (linear regression, r = 0.96): the larger the dead-space volume of the filter, the more RBCs and hemoglobin lost.

Filtration at 4°C or RT did not significantly affect the volume lost with the same filter. All filters produced filtered RBCs with various volumes (p < 0.001 for all filters, except when stated otherwise in the legend of Table 3), and, consequently, the number and volume of RBCs and of the hemoglobin lost during filtration were also significantly different.

The BPF4 was the smallest filter tested, with a volume loss of only 22 ± 1 mL, reflected as an RBC loss of 0.14 ± 0.02 x 10¹² (13 ± 2 mL) cells and a hemoglobin loss of 4.3 ± 1.1 g.

The Optima Plus, the largest filter in these experiments, yielded 3 filtered RBC units with a volume of RBCs below our specifications (i.e., <119 mL), while 5 filtered RBC units had a low (<38 g) hemoglobin content. Leucoflex, the second largest filter, produced 1 unit with a low hemoglobin content. All other units conformed to our specifications.

**Filtration time**

The Sepacell and the BPF4 were filled by squeezing the RBC bag. Filling occurred within 15 seconds at RT and took slightly longer at 4°C, as shown in Table 4. All other filters were filled spontaneously, taking from about 38 seconds for the Imugard up to more than 7 minutes for the Optima Plus. Filling at 4°C took longer to complete than that at RT (p < 0.0001, paired t test). The quickest filtration was achieved with the BPF4 at RT, in an average of 6 minutes.

**Influence of cooling and warming on WBCs in BC-depleted RBC concentrates**

To exclude the possibility that WBCs would fragment because of the quick cooling, resulting in an apparently low WBC content after cooling, we performed WBC counts before and after cooling and after rewarming. Before cooling, the mean WBC concentration in 9 BC-depleted RBC concentrates was 1.9 ± 1.3 x 10⁶ per mL. After cooling to 4°C, the number of WBCs was slightly but significantly higher: 2.0 ± 1.3 x 10⁶ per mL (p = 0.02, paired t test). After rewarming, the WBC concentration was again 1.9 ± 1.3 x 10⁶ per mL. We presume that the slightly higher number of WBCs after cooling is due to the counting of platelet aggregates as WBCs by the cell analyzer. From these data, it can be concluded that quick cooling did not have a diminishing effect on the WBC content.

**DISCUSSION**

In this study, we evaluated the performance of six WBC-reduction filters for BC-depleted RBC concentrates. All tested filters could remove WBCs to a level below the current upper limit for WBC reduction of 5 x 10⁶ per unit. The best results were obtained with Leucoflex, BPF4, Sepacell (all three at 4°C), and Imugard (at RT) filters. The performance at RT of Imugard, a polyurethane filter, was as good as that of the polyester filters at 4°C. We found that, with polyester filters, filtration at 4°C provided the best results. When 2 identical units were filtered at both RT and 4°C, the number of residual WBCs at RT was at least double the number found at 4°C.

In polyester filters, capture of WBCs is caused by three mechanisms: 1) mechanical sieving; 2) direct adhesion of WBCs to the filter material; and 3) indirect adhesion of WBCs to adhering platelets. Filtration of WBCs is a combined result of these mechanisms, which are in addition temperature-dependent.

As shown by Steneker et al., activated platelets and platelets with pseudopodia play a major role in indirect granulocyte adhesion, whereas lymphocytes are mainly captured by mechanical sieving. Because the WBCs present

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**TABLE 3. Volume and RBC measures of filtered RBCs, after filtration of BC-depleted RBC concentrates with comparable composition, as obtained by pooling and dividing**

<table>
<thead>
<tr>
<th>Filter</th>
<th>Leucoflex (4°C)</th>
<th>Leucoflex (20°C)</th>
<th>Cellselect FR (4°C)</th>
<th>Cellselect FR (20°C)</th>
<th>Optima Plus (4°C)</th>
<th>Optima Plus (20°C)</th>
<th>BPF4 (4°C)</th>
<th>BPF4 (20°C)</th>
<th>Sepacell (4°C)</th>
<th>Sepacell (20°C)</th>
<th>Imugard (4°C)</th>
<th>Imugard (20°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume (mL)</td>
<td>227 ± 9</td>
<td>240 ± 9</td>
<td>235 ± 9</td>
<td>213 ± 9</td>
<td>251 ± 8</td>
<td>252 ± 7</td>
<td>244 ± 6</td>
<td>243 ± 7</td>
<td>234 ± 6</td>
<td>234 ± 6</td>
<td>234 ± 6</td>
<td>234 ± 6</td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>56.8 ± 0.9</td>
<td>56.8 ± 0.9</td>
<td>56.8 ± 0.9</td>
<td>57.8 ± 0.9</td>
<td>58.8 ± 0.9</td>
<td>58.8 ± 0.8</td>
<td>58.8 ± 1.2</td>
<td>58.3 ± 1.1</td>
<td>58.8 ± 0.6</td>
<td>58.8 ± 0.6</td>
<td>58.8 ± 0.6</td>
<td>58.8 ± 0.6</td>
</tr>
<tr>
<td>RBC volume (mL)</td>
<td>132 ± 7</td>
<td>142 ± 7</td>
<td>138 ± 7</td>
<td>123 ± 6</td>
<td>148 ± 6</td>
<td>147 ± 6</td>
<td>143 ± 6</td>
<td>142 ± 6</td>
<td>137 ± 5</td>
<td>137 ± 5</td>
<td>137 ± 5</td>
<td>137 ± 5</td>
</tr>
<tr>
<td>RBC count (10¹²)</td>
<td>1.49 ± 0.08</td>
<td>1.56 ± 0.07</td>
<td>1.53 ± 0.07</td>
<td>1.37 ± 0.07</td>
<td>1.64 ± 0.07</td>
<td>1.64 ± 0.06</td>
<td>1.59 ± 0.06</td>
<td>1.58 ± 0.07</td>
<td>1.52 ± 0.05</td>
<td>1.52 ± 0.05</td>
<td>1.52 ± 0.05</td>
<td>1.52 ± 0.05</td>
</tr>
<tr>
<td>Hemoglobin (g)</td>
<td>41.5 ± 2.5</td>
<td>44.4 ± 2.1</td>
<td>43.8 ± 2.1</td>
<td>39.0 ± 1.9</td>
<td>46.5 ± 2.0</td>
<td>46.6 ± 2.2</td>
<td>45.2 ± 2.1</td>
<td>45.1 ± 1.9</td>
<td>43.1 ± 1.9</td>
<td>43.1 ± 1.9</td>
<td>43.1 ± 1.9</td>
<td>43.1 ± 1.9</td>
</tr>
</tbody>
</table>

* Results reported as mean ± SD (n = 12).
† All differences p < 0.001, except B vs. C, G, and H (NS); C vs. I (NS); E vs. F (NS); G vs. H (NS); and I vs. A and B (p < 0.01).
‡ All differences except D vs. B, E, F, G, and I (p < 0.01).
§ All differences p < 0.001, except B vs. G and H (NS); C vs. I (NS); E vs. F (NS); G vs. H (NS); B vs. C and I (p < 0.01); C vs. H (p < 0.01); and F vs. G and I (p < 0.01).
¶ All differences p < 0.001, except B vs. C, G, and H (NS); C vs. I (NS); E vs. F (NS); G vs. H (NS); B vs. I (p < 0.05); E vs. G (p < 0.05); F vs. G (p < 0.01); and H vs. I (p < 0.01).
‖ All differences p < 0.001, except B vs. C, G, H, and I (NS); C vs. I (NS); E vs. F, G, and H (NS); G vs. H (NS); C vs. H (p < 0.05); F vs. G (p < 0.05); A vs. I (p < 0.01); C vs. G (p < 0.01); and F vs. H (p < 0.01).
TABLE 4. Mean ± SD filling and total filtration time of BC-depleted RBC concentrates (n = 12) with comparable composition, as obtained by pooling and dividing

<table>
<thead>
<tr>
<th>Filter</th>
<th>Filling Time (min:sec)</th>
<th>Total Filtration Time (min:sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leucoflex (4°C)</td>
<td>A: 6:16 ± 1:02</td>
<td>35:59 ± 2:46</td>
</tr>
<tr>
<td>Cellselect FR (20°C)</td>
<td>B: 1:04 ± 0:06</td>
<td>10:34 ± 1:55</td>
</tr>
<tr>
<td>Cellselect FR (4°C)</td>
<td>C: 2:30 ± 0:17</td>
<td>10:35 ± 3:12</td>
</tr>
<tr>
<td>Optima Plus (20°C)</td>
<td>D: 7:07 ± 2:41</td>
<td>16:39 ± 3:37</td>
</tr>
<tr>
<td>BPF4 (20°C)</td>
<td>E: 1:11 ± 0:02</td>
<td>8:00 ± 1:03</td>
</tr>
<tr>
<td>BPF4 (4°C)</td>
<td>F: 0:22 ± 0:03</td>
<td>10:51 ± 0:53</td>
</tr>
<tr>
<td>Sepacell (20°C)</td>
<td>G: 0:12 ± 0:02</td>
<td>7:47 ± 0:29</td>
</tr>
<tr>
<td>Sepacell (4°C)</td>
<td>H: 0:18 ± 0:13</td>
<td>13:53 ± 1:13</td>
</tr>
<tr>
<td>Imugard (20°C)</td>
<td>I: 0:38 ± 0:04</td>
<td>6:57 ± 0:41</td>
</tr>
</tbody>
</table>

* All differences p<0.001, except B vs. F and H (NS); E vs. G and I (NS); G vs. I (NS); D vs. H (p<0.05); F vs. G (p<0.05); B vs. D (p<0.01); and F vs. H (p<0.01).

in BC-depleted RBC concentrates are mainly granulocytes, the number of platelets present is of importance for filter performance. This hypothesis has been supported by later studies, in which it was established that the presence of high numbers of platelets in non-BC-depleted RBC concentrates resulted in better WBC reduction than that in BC-depleted RBC concentrates. Although a bedside filter was used in one study, the filter performance was markedly decreased when the platelet load was less than 100 x 10^6.

Further, we conjecture that direct and indirect adhesion occurs mainly at RT, when the membrane fluidity of the cells enables direct adherence. Membrane fluidity permits the WBCs to pass through pores much smaller than the actual cell diameter, and, therefore, sieving will play a relatively small role in the filtration process at RT. In contrast, we hypothesize that, at 4°C, mechanical sieving will be the most prominent mechanism of WBC removal, because membrane rigidity prevents cells from passing through small pores. In our experiments and those of others, filtration at RT was less efficient than filtration at 4°C. We believe that this was true because the adhesion of WBCs was not able to compensate for the loss of mechanical sieving. Filtration at 4°C has the consequence that, according to current Good Manufacturing Practices, the units must be cooled in a controlled manner, and the filtration must be performed in a refrigerated room to satisfy the strict temperature control. This requirement may, however, have some logistical disadvantages.

The better performance at RT of the Imugard (polyurethane filter) than of the polyester filters may be due to the filtration material: it has been stated that the pores of polyurethane can be made so small that WBCs are retained, even at RT. In that case, mechanical sieving is the most significant filtration mechanism; adhesion to the filter material plays an insignificant role.

Another possible contribution to the filtration performance may be the flow rate. At 4°C, blood viscosity is higher, resulting in a decreased flow rate, which may enhance the probability of platelet and WBC capture in the filter material. A study using bedside filtration showed that filtration at 4°C gave better WBC reduction than that at RT, even when the flow rate was equal for both; this suggested that other temperature-dependent mechanisms, such as sieving, are present.

The age of the RBCs at the time of filtration is an additional factor that should be considered. In our blood bank, RBCs are routinely filtered within 24 hours after phlebotomy. Our consideration is that WBCs deteriorate quickly after phlebotomy and, because polyester filters do not adequately remove WBC fragments, HLA-bearing particles may still be present in the filtered RBCs, although no intact WBCs can be counted. As hypothesized by Sintnicolaas et al., secondary HLA alloimmunization may still occur, because of the presence of microparticles. However, to address this possibility, further in vitro and in vivo studies should be performed.

The current standard for WBC-reduced components is less than 5 x 10^6 WBCs per unit, but the Council of Europe recently recommended that WBC-reduced blood components should not contain more than 1 x 10^6 WBCs per unit. Only Leucoflex, BPF4, Sepacell (all at 4°C), and Imugard (at RT) conform to this more stringent demand for all tested units.

We conclude that all filters conform to the current standards for WBC reduction. For routine preparation of WBC-reduced RBCs, the composition, temperature, and age of the RBCs and the logistic preferences should be taken into account in the final choice of a filter. Moreover, it is of great importance to validate the chosen filter under routine conditions before implementation.

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