Pooled platelet concentrates prepared by the platelet-rich plasma (PRP) method were filtered with three different filters and stored for 8 days at room temperature. The effect of filtration on leukocyte contamination, platelet concentration, and the in vitro function, morphology, metabolism and activation of platelets were studied. Eight pools of 20 PRP-PC were used, each pool was split into 4 equal volumes; 3 were filtered over a PL50HF, a PL-10A and a Bio PIO filter, the 4 served as a control. After filtration, leukocyte counts exceeded 3 x 10^4 in none of the pooled PC. Platelet loss induced by filtration was about 17%. During storage, no differences in pH, PCO₂, and lactate and glucose concentration were found between the filtered and the unfiltered units, nor were any differences observed between filtered and unfiltered pooled PC in aggregation upon stimulation with collagen and/or ADP, adhesion capacity to collagen in flowing blood, nucleotide content of the platelets and nucleobase concentration in the plasma, expression of activation-dependent antigens, or platelet morphology as observed by light microscopy and by the swirling effect. Selective removal of β-thromboglobulin (22%) by the PL50HF filter was observed. Pooled PC prepared by the PRP-method can be filtered and stored for 8 days without detrimental effect on platelet function, metabolism or activation.

Introduction

Donor leukocytes in platelet transfusion can cause adverse effects in the recipient: alloimmunization against leukocyte (HLA) antigens, nonhemolytic febrile transfusion reactions, refractoriness to platelets [1, 2] and transmission of certain infectious agents [3]. To prevent or reduce these post-transfusion complications, donor leukocytes can be removed from the platelet concentrate (PC) by filtration. The absolute number of leukocytes that may be left in the PC without causing adverse effects is not clear yet [4], but a maximal reduction of leukocytes has to be pursued with an acceptable loss of platelets. However, the number of contaminating leukocytes after filtration will also depend on the number and nature of the leukocyte present in the PC before filtration. In an earlier study we filtered pooled PC prepared by the buffy coat (BC) method with a total leukocyte contamination of 7.2±1.8x10^4, resulting in a post-filtration contamination of less than 5x10^4 in all PC [5, 6]. Outside Europe, where most PC are prepared by the BC method, the number of contaminating leukocytes is lower, generally less than 5x10^4 in all PC.
method, PC are usually prepared by the platelet-rich plasma (PRP) method. PC prepared by the PRP method have a higher leukocyte contamination [7, 8] than PC prepared by the BC method [9], which may lead to higher post-filtration leukocyte contamination. Moreover, the composition of leukocyte contamination is different, with a large amount of granulocytes in PRP-PC, whereas mainly mononuclear cells are found in the BC-PC. Platelets prepared by the PRP method are more activated immediately after preparation [10], which may result in higher platelet loss during filtration, whereas also the higher red cell contamination may influence filtration results. The more activated state of platelets prepared by the PRP method may also influence the effect of filtration on the in vitro platelet function during long-term storage after filtration. No effect of filtration on platelet function and activation was found before [11-14], but it was found that after filtration the percentage platelets expressing CD62 was reduced [15].

Materials and Methods

Blood Collection and PC Preparation
Platelet concentrates were prepared from 500 ml of blood collected in 70 ml citrate-phosphate-dextrose (CPD) in PVC/DEHP quadruple systems (NPBI BV, Emmer-Compascuum, the Netherlands). After storage at 20±2°C for 12-16 h [16], the blood was separated in red cells and PRP by centrifugation for 7 min at 1,000 g at 20°C. The PRP was pressed with reduced flow into an empty satellite bag using an automated device for component preparation, the Compomat with Compo-master software [17] (NPBI). The PRP was subsequently centrifuged for 6 min at 3,000 g at 20°C. The Compomat was then used with a different program to remove the plasma until about 70 ml plasma was left. The platelet pellet with the 70 ml plasma was left undisturbed for 90 min at room temperature before manual resuspension. Subsequently, the PC were stored overnight in 600 ml PVC/DEHP bags, in a platelet incubator (22±1°C) on a horizontal flat bed shaker (1 cycle/s) (Helmer labs inc, Noblesville, Ind., USA) before pooling.

Twenty PC of the same ABO blood group (A positive) were pooled in a 2,800 ml PVC bag (Biotrans, Emmer-Compascuum, the Netherlands) using a sterile connecting device (SCD 312, Haemonetics, Braintree, Mass., USA). The PC were selected by platelet count leading to a final platelet concentration that would be similar for all 8 pools used in this study. Each pool was split into 4 units with identical volume. One unit (control pool) was transferred to a 1,000 ml polylefin storage bag (NPBI), the other 3 in 600 ml PVC transfer bags (NPBI).

Filtration of Platelet Concentrates
Three leukocyte depletion filters for platelets were used: the PL50HF (Pall Biomed. Products Co., Glen Cove, N.Y., USA); the PL-10A (Sepacell, Asahi Medical Co., Tokyo, Japan) and the BioP10 (Biofil, Italy). The PC and the bag for storage of the platelets after filtration (1,000 ml polylefin) were connected by SCD to the respective filters with tube lengths according to manufacturer's instructions. Filtration with the PL50HF and the Biofil filter were performed at gravity flow rate (resulting in a flow rate of approximately 100 ml/min and 60 ml/min, respectively), with the PL-10A the flow was adjusted to 20 ml/min, according to the manufacturer's instructions. The platelet concentration and volume of the control pool was adjusted to the mean platelet concentration and volume of the filtered pooled PC after filtration, by adding plasma and subsequently removing platelet-rich plasma.

The PC were stored for 8 days in the platelet incubator (22±1°C, 1 cycle/s). The total storage time was 12-16 h as whole blood. 1 day as single PC and 7 days as pooled PC. Samples were taken aseptically via a sampling site coupler (NPBI) on day 1, 2, 3, 6 and 8.

Swirling Patterns and Platelet Morphology
Swirling patterns were determined during storage by one person throughout the whole study and scored according to Fratantoni [18]. To evaluate morphology 50 µl of a platelet sample was fixed with 250 µl 0.5% glutaraldehyde in phosphate-buffered saline (PBS) at room temperature and stored for evaluation at 4°C. The morphology was judged by light microscopy (Leitz, Wetzlar, Germany) with oil immersion (1,000×), using a modification of the Kunicki score [19], with a maximal score of 400. In this modification in a 100 cell count the number of discs was multiplied with 4, the number of dendrites with 2 and the number of spheres with 1.

Platelet Adhesion
Platelet interaction with collagen in flowing blood was studied in a rectangular perfusion chamber as described before [5].

Platelet Aggregation
Platelet aggregation was measured in a lumiaggregrometer (Lumiaaggregation module, model 100B, Payton, Buffalo, N.Y., USA) as described before [20]. All samples were diluted with plasma (blood group A, Rh D positive) until a platelet concentration of 250×10⁶/l was obtained. Aggregating agents used were collagen (Kollagen Regent Horm., Hormon Chemie) in a concentration of 6.7 µg/ml and collagen plus ADP (adenosine 5-diphosphate, sodium salt. Sigma Chem. Co, St. Louis, Mo., USA) in the double stimulus mixture in a concentration of 11 µM and 2.2 µg/ml, respectively.

Nucleotide Content and Nucleobase Release
The nucleotide content of platelets was analyzed with an anion-exchange HPLC method after perchloric acid extraction as described before [21]. The neutralized samples were kept at -70°C until HPLC analysis. Columns were prepurified Partisphere Partisol-5 SAX cartridges (125×4.5 mm internal dimensions, Whatman, Chifton, N.Y., USA).

Nucleobase release by the platelets as degradation products of nucleotides was measured. Analysis of hypoxanthin, xanthin, uric acid, adenine and adenosine in perchloric acid extracts of supernatants were performed by reversed-phase HPLC according to van Gennip et al. [22]. The C18 columns used were Partisphere 5 C18 (125×4.5 mm internal dimensions, Whatman). The concentrations of the different substances were calculated by comparison with highly purified standards (Sigma Chemical Co).

Activation Dependent Antigens
To measure activation of platelets, murine monoclonal antibodies (McAbs) directed against human platelet antigens were used as described before [23]. In addition to the earlier described McAb C17 and McAb Y2 against GP IIa alone or in complex with GP IIb, McAb C8 against GMP 140 and McAb 435 against a 53 kD protein. McAb 45
β-Thromboglobulin Release and Protein Concentration

The release of β-TG and the total protein content in the supernatant of platelets was measured after centrifugation for 5 min at 12,000 g and storage of the samples at -80°C until the measurements were performed. β-TG was measured with a RIA (Amersham, Little Chalfont, UK). The protein concentration was measured with a BCA (bicinchoninic acid) Protein Assay kit (Pierce, Rockfield, Ill., USA).

Other in vitro Parameters

Volumes of the pooled PC were calculated by dividing the net weight of the bag contents by its specific gravity. Platelet counting was performed on a Cell-dyn 100 whole blood platelet counter (Sequana-Turner Co, Mountain View, Calif., USA). Leukocytes were counted electronically with a Coulter Multisizer II (Coulter Electronics, Mijdrecht, the Netherlands) or, after filtration, by fluorescence light microscopy in a Nageotte bright-line haemocytometer (Superior, Bad Mergentheim, Germany). For this measurement 100 μl PC was mixed with 400 μl acridine orange solution (0.05 mg/ml in PBS), incubated for a few minutes at room temperature after which the Nageotte chamber was filled. After 15 min settling in a humidified atmosphere, the number of leukocytes in 2 compartments of 50 μl of the Nageotte hemocytometer were counted. Under these conditions 1 counted cell represents a leukocyte concentration of 50 cells/ml.

The amount of free hemoglobin in the PC (after addition of some lysis fluid) was measured spectrophotometrically (Spectronic 301, Milton Roy, Rochester, N.Y., USA), using the absorption at 415 nm corrected for the color of plasma. The hemoglobin content was used to calculate the number of contaminating erythrocytes.

Glucose and lactate were measured enzymatically in the supernatant after centrifugation of a PC sample [25], pH, PO2, and PCO2 were measure at 37°C on a 238 pH/Blood gas analyzer (Ciba Corning, Essex, UK).

Statistical Evaluation

Statistical analysis was performed with the Student t test for unpaired and paired observations and differences were considered significant at p<0.05. When appropriate, significance levels were adjusted for multiple comparison according to the method of Bonferroni-Holm [26]. The total experiment-wise error was chosen to be lower than 0.05. For the statistical analysis of changes during storage for each pooled PC type, only the results on day 1 and day 8 were compared.

Results

Platelet Concentrates

In table 1 the volume, platelet content and leukocyte contamination of the pools of PC before and after filtration and of the control pool are shown. The mean erythrocyte contamination after filtration was 5±1.3×10^6. Filtration with the PL50HF filter caused a platelet loss of 13%, the PL-10A filter of 17% and the Bio P10 of even 20% (NS) due to loss of volume in the tubing and the filter and trapping of platelets in the filter. All filter effectively removed leukocytes from the pooled PC to a postfiltration leukocyte contamination of maximal 0.3×10^6. After filtration of the pool with a leukocyte contamination of 71±16.1×10^6 by the PL-10A filter, leukocytes were reduced by >99.9% (in two samples no cells were observed in the Nageotte chamber) to 0.03±0.03×10^6 leukocytes (range <0.02–0.10×10^6). This is significantly lower (p<0.005) than after filtration by the PL50HF filter (99.8±0.11%, range 0.02 to 0.24×10^6 leukocytes), or after filtration by the Bio P10 filter (99.8±0.12%, range 0.03 to 0.30×10^6 leukocytes) (table 1).

In table 2 the pH, PCO2, PO2 and swirling and morphology scores of the filtered and unfiltered pooled PC during the 8-day storage period are shown. During storage, the normal changes in pH, PCO2 and PO2 were found with a pH well above 6.8 on day 8 of storage. Although some significant differences were found, i.e. a significantly lower pH in the pooled PC filtered by the PL50HF filter on all storage days; a higher PCO2 in the same pooled PC on day 1 of storage and a significantly higher PO2 in the pooled PC filtered by the PL-10A filter on day 6 (not shown) compared to the control pool and the pooled PC filtered by the Biofil filter, these differences are too small to influence the platelet quality during storage. Swirling on day 1 was maximal in all pooled PC, filtered or unfiltered and decreased during storage but was still good at day 8, with a range from 1.5 to 2 in all grouped PC; no differences were observed between the various pooled PC. Morphology scores were excellent on day 1 (at least 300 in all pooled PC, with a mean percentage of discs of 89%), with no significant differences between the pooled PC and decreased to about 200 on day 8 which is still acceptable, although in a few pooled PC in all groups the morphology score dropped below 200. Bizarre shapes as described by Shimizu et al. [27] during storage of platelets in polyolefin containers were also observed in an increasing percentage during storage.

Glucose Consumption and Lactate Production

No significant differences were observed between the various pooled PC for the glucose consumption and lactate production during the 8-day storage period (table 3). The ratio lactate production/glucose consumption was below 2, indicating oxidative metabolism of glucose throughout storage.

Nucleotide Content and Nucleobase Release

No significant differences in nucleotide content or nucleobase release between the pooled PC filtered over the
Table 1. Volumes, leukocyte and platelet concentration in the pooled PC before and after filtration with the PL50HF, the PL-10A and the Bio P10 filter and in the non-filtered control pool

<table>
<thead>
<tr>
<th></th>
<th>Before filtration</th>
<th>Pall PL50HF</th>
<th>Sepacell 10A</th>
<th>Bio P10</th>
<th>Control Volume adjusted</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume (ml)</td>
<td>331±17.5</td>
<td>321±15.3</td>
<td>313±16.1</td>
<td>314±22.1</td>
<td>313±18.1</td>
</tr>
<tr>
<td>Platelets, × 10⁶</td>
<td>376±57.2</td>
<td>325±35.6</td>
<td>301±12.3</td>
<td>296±30.5</td>
<td>310±44.6</td>
</tr>
<tr>
<td>WBC, × 10⁶</td>
<td>71±16.1</td>
<td>0.12±0.077</td>
<td>0.03±0.035</td>
<td>0.12±0.089</td>
<td>56.2±18.00</td>
</tr>
</tbody>
</table>

Shown are the mean ± SD, n = 8.

Table 2. pH, PO₂, PCO₂, and platelet morphology in the filtered and non-filtered control pools during storage

<table>
<thead>
<tr>
<th>Day</th>
<th>Pall 50HF</th>
<th>Sepacell 10A</th>
<th>Bio P10</th>
<th>Control</th>
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<td></td>
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<tr>
<td>1</td>
<td>7.10±0.021</td>
<td>7.14±0.021</td>
<td>7.13±0.023</td>
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</tr>
<tr>
<td>3</td>
<td>7.11±0.018</td>
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</tr>
<tr>
<td>8</td>
<td>6.93±0.023</td>
<td>6.95±0.021</td>
<td>6.96±0.029</td>
<td>6.97±0.044</td>
</tr>
<tr>
<td></td>
<td>59±3.9</td>
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<td>56±4.4</td>
<td>57±3.6</td>
</tr>
<tr>
<td></td>
<td>52±3.4</td>
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<td>50±3.4</td>
<td>50±2.3</td>
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<tr>
<td></td>
<td>49±2.9</td>
<td>48±2.9</td>
<td>48±3.2</td>
<td>46±2.1</td>
</tr>
<tr>
<td></td>
<td>59±3.9</td>
<td>53±3.7</td>
<td>56±4.4</td>
<td>57±3.6</td>
</tr>
<tr>
<td></td>
<td>52±3.4</td>
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<td>50±3.4</td>
<td>50±2.3</td>
</tr>
<tr>
<td></td>
<td>49±2.9</td>
<td>48±2.9</td>
<td>48±3.2</td>
<td>46±2.1</td>
</tr>
<tr>
<td></td>
<td>117±16.6</td>
<td>128±9.2</td>
<td>109±10.5</td>
<td>103±5.0</td>
</tr>
<tr>
<td></td>
<td>93±6.0</td>
<td>94±6.3</td>
<td>98±7.9</td>
<td>97±4.3</td>
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<tr>
<td></td>
<td>102±8.2</td>
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<td>107±5.9</td>
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<tr>
<td></td>
<td>3.0±0.00</td>
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<td>3.0±0.00</td>
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</tr>
<tr>
<td></td>
<td>2.5±0.38</td>
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</tr>
<tr>
<td></td>
<td>1.8±0.27</td>
<td>1.8±0.27</td>
<td>2.0±0.00</td>
<td>2.0±0.00</td>
</tr>
<tr>
<td></td>
<td>344±17.8</td>
<td>338±13.1</td>
<td>334±15.5</td>
<td>351±17.2</td>
</tr>
<tr>
<td></td>
<td>292±23.7</td>
<td>298±22.8</td>
<td>293±18.1</td>
<td>306±12.7</td>
</tr>
<tr>
<td></td>
<td>202±20.0</td>
<td>203±7.1</td>
<td>193±11.0</td>
<td>200±18.5</td>
</tr>
</tbody>
</table>

Shown are the mean ± SD, n = 8.

Morphology was scored according to Kunicki with a maximal score of 400.

From the degradation products of purine nucleotides, only hypoxanthin increased during storage from a mean of 9 ±3.6 pmol/10⁶ platelets on day 1 to 54±15.0 pmol/10⁶ platelets on day 8, whereas minimal changes were found in xanthin and uric acid concentration (not shown). Small amounts of adenine (up to 40 μM) were initially present in the pooled PC (not shown), due to contamination of the plasma with some erythrocyte additive solution (containing 1.25 mM adenine), but after 8 days of storage only in some pooled PC traces of adenine were detectable. Adenosine was below the detection limit of 50 nM.

β-Thromboglobulin Release

After filtration over the PL50HF filter β-TG was significantly lower (22%) than in the pooled PC filtered over the other two filters (table 3). The total protein concentration of the supernatants from day 1 were similar, suggesting a selective removal of β-TG by the PL50HF filter, like found before [5]. The values of the filtered pooled PC cannot be compared with the control pool because, due to the adjustment for platelet concentration and pool-volume with plasma with a low β-TG concentration, this control pool had a different composition. During storage, a similar increase of the amount of released β-TG was found for all pooled PC.
Fig. 2. Adhesion to collagen of filtered and subsequently stored platelets. Platelet adhesion (mean ± SD) to collagen, expressed as the percentage of coverage. Platelets were stored for 8 days after filtration by the PL50HF filter (Δ-Δ), the PL-10A filter (○-○) or the Bio P10 filter (■-■); the control platelets (---) were not filtered (n = 8).

Fig. 3. Effect of filtration on platelet activation, immediately after filtration and during subsequent storage. Platelet activation was measured by binding with McAb. For McAb C17 (a), against GP IIb-IIIa, the mean (±SD) fluorescence intensity (MFI) was used. For McAb C8 (b), against GMP 140, and McAb 435 (c), against a 53-kD protein, the percentage of positive cells (mean±SD) above the negative control was used. Platelets were stored for 8 days after filtration by the PL50HF filter (triangle), the PL-10A filter (●-●) or the Bio P10 filter (■-■); the control platelets (---) were not filtered (n = 8).
Table 3. Nucleotide content, glucose consumption, lactate production and β-thromboglobulin release in the filtered (with the PL50HF, the PL-10A and the Bio P10 filter) and non-filtered control pools during storage.

<table>
<thead>
<tr>
<th></th>
<th>Day</th>
<th>Pall 50HF</th>
<th>Sepacell 10A</th>
<th>Bio P10</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose¹</td>
<td>1-8</td>
<td>0.6±0.06</td>
<td>0.7±0.07</td>
<td>0.7±0.06</td>
<td>0.6±0.09</td>
</tr>
<tr>
<td></td>
<td>1-8</td>
<td>1.0±</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adenylc acid³</td>
<td>1</td>
<td>58±8.3</td>
<td>60±9.0</td>
<td>59±6.5</td>
<td>58±4.7</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>59±7.3</td>
<td>60±6.7</td>
<td>59±2.5</td>
<td>56±6.2</td>
</tr>
<tr>
<td></td>
<td>8</td>
<td>43±3.8</td>
<td>43±4.3</td>
<td>46±3.7</td>
<td>42±3.3</td>
</tr>
<tr>
<td>ATP⁴</td>
<td>8</td>
<td>81±7.4</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

β-TG⁵

Shown are the mean ± SD, n = 8.

¹ Consumption in nmol/10⁶ platelets/day.
² Production in nmol/10⁶ platelets/day.
³ Adenylc acid = AMP + ADP + ATP content in pmol/10⁶ platelets.
⁴ Expressed as % left from day 1.
⁵ β-TG released by the platelets in the plasma, in ng/10⁶ platelets.

Fig. 1. Platelet aggregation after filtration and during storage. Platelet aggregation (mean ± SD) upon stimulation with 40 μM ADP (lower 4 lines) and 11 μM ADP and 2.2 μg/ml collagen as a double stimulus (upper 4 lines). Platelets were stored for 8 days after filtration by the PL50HF filter (▲▲), the PL-10A filter (●●) or the Bio P10 filter (■■); the control platelets (---) were not filtered (n = 8).

Platelet Adhesion
No significant differences in the adhesion to collagen (fig. 2) were observed between the 4 different types of pooled PC either immediately after filtration or during storage. Over the 8 day storage period a slight but significant decrease in the adhesion to collagen was found.

Activation Dependent Antigens
The binding of McAb C17 (fig. 3a) against the GP IIb-IIIa complex increased during storage, especially between day 1 and 3, whereas binding of McAb Y2 also increased during storage but to a lesser extent (not shown). No significant differences were found in GP (IIb-IIIa) expression be-
Discussion

In an earlier study [5, 6] we filtered PC prepared from pooled BC with a leukocyte contamination of 7.2 ± 1.8 x 10^6, resulting in a postfiltration contamination of less than 5 x 10^4, which is probably below the minimal number of leukocytes that can cause alloimmunization [28, 29]. In this study we used a pool of 5 PC, prepared by the PRP method which results in a higher leukocyte contamination after filtration compared to the BC method. However, the total initial leukocyte contamination (71 ± 16.1 x 10^6) and the total initial erythrocyte contamination (5.2 ± 1.30 x 10^7) were not as high as is often found in PRP-PC [7, 8], probably due to the fact that our PC were prepared in a standardized way with flow reduction on an automated device, resulting in a somewhat lower platelet recovery but also in a lower leukocyte contamination. Another difference with the usual way of preparation is that we introduced a storage period of 12–16 h at 20°C before component preparation [16]. However, in our hands, hardly no difference was found in leukocyte contamination and platelet concentration when PRP was prepared within 6 h after blood collection (data not shown). Although all filters effectively removed leukocytes from the pooled PC, after filtration leukocytes could still be detected in the Nageotte counting chamber in contrast to the situation when BC-PC were filtered by the Pall or the Sepacell filter [6]. The postfiltration leukocyte contamination of the PRP-PC did not exceed 3 x 10^5, which is also below the suggested limit of the number of leukocytes which can cause alloimmunization [28, 29], but when PRP-PC with a higher leukocyte contamination are filtered, post-filtration leukocyte contamination may increase.

Platelet loss was 13–20% depending on the type of filter, which is quite high, as most filtration studies report a platelet loss of 5–20% [12, 13, 30]. The platelet loss can not be explained by loss of volume in the filter and in the tubing alone because the platelet concentration also decreased, indicating trapping of platelets in the filters. Platelets prepared by the PRP method are more activated immediately after preparation compared to BC-PC [10], which was confirmed in this study. In a study by Pedigo et al. [15], it was found that after filtration of activated platelets, expressing the activation marker CD62, platelet loss was positively correlated with the percentage CD62-positive cells, due to selective trapping of CD62-positive platelets in the filter. This mechanism cannot explain the platelet loss in our study because no difference in the amount of CD62-positive platelets was found between the filtered PC and the unfiltered control. However, our starting value of CD62-positive platelets was considerably lower than found by Pedigo (although higher than in BC-PC [5]), which may be due to the holding period of the whole blood, prior to component preparation. Both after filtration of BC-PC [6] or PRP-PC (this study) over a PL50HF filter a 10% decrease of platelet concentration was observed, while after filtration over the Sepacell PL-10A filter the decrease in platelet concentration was 10% for BC-PC and 16% for PRP-PC. This suggests that there is a difference in sensitivity between the 2 filters for the activation state of the platelets, although clearly not correlated to CD62 expression.

After filtration of PC prepared by the BC method no effect of filtration on platelet metabolism, activation or in vitro function was found [5, 6]. The increased activation state of PRP platelets could also influence the effect of filtration on the platelets. However, again no differences between the filtered pooled PC and the unfiltered control pool in the expression of activation antigens, platelet aggregation, platelet adhesion to collagen in flowing blood, platelet nucleotide content or nucleobase release, platelet morphology or platelet glucose consumption and lactate production were found. This is in agreement with other studies in which no effect of filtration on various in vitro and in vivo parameters of platelets prepared by the PRP method was found [11–14], however in these studies PC were not stored after filtration.

Depletion of leukocytes from PC might also improve the quality of platelets during storage because leukocytes have a detrimental effect on platelets during storage [31, 32]. However, no positive effect of leukocyte reduction on platelet function was observed, as was found also for the BC-PC (with a 10-fold lower initial leukocyte contamination). This might be explained by the fact that all PC were stored under optimal conditions in a polyolefin container with high gas exchange capacity, which might prevent a pH drop caused by contaminating leukocytes and by the fact that the prefiltration leukocyte contamination was not high enough to cause adverse effects on platelet function [32].

We conclude that a pool of PC prepared by the PRP method can be filtered and stored for 8 days at room temperature, without detrimental effect on platelet function, metabolism or activation. In countries where the PRP method of PC preparation is used, these PC, with a leukocyte contamination of less than 3 x 10^6 per unit, might be of clinical importance for the treatment of patients by transfusion of platelets with good in vitro parameters and a minimal risk for the development of HLA immunization.
References


