We have previously shown that donation of blood into anticoagulants containing half the normal amount of citrate results in a dramatic improvement in the stability of coagulation factor VIII and has no adverse effect on the in vitro qualities of red cells or platelets during storage. To confirm the viability of stored cellular components we are now performing autologous survival studies in healthy volunteers using radiolabelled cells from red cells and platelets stored for 35 and 5 days respectively. Results to date indicate a 24 hour survival of 80% for red cells stored at a haematocrit of 0.70 for 30 days. Infusion of III-In oxine labelled platelets after storage for 5 days in full or half-strength citrate gave recoveries of 40% and survival of 7 days. These encouraging results suggest use of half-strength without any additional donor recruitment. Further in vitro studies have also been performed on cellular components and reveal survival studies in healthy volunteers using radiolabelled cells or platelets during storage. To confirm the viability of red cells stored at a haematocrit of 3. 

Platelets are routinely stored for transfusion at room temperature in autologous, citrated plasma. We have demonstrated previously that these conditions do not completely block activation of plasma enzyme systems, as indicated by generation of thrombin activity (Vox Sang, 51:192, 1986). Here, we demonstrate the conversion of large amounts of complement factor C3 during storage of citrated PC by using radioimmunoassay quantitation of the activation peptide C3a des-Arg (Upjohn Diagnostics). Supernatant samples from stored PC and from citrated platelet-poor plasma (PPP) stored under the same conditions showed a rapid linear increase in C3a levels over time with no significant difference (paired t-test, p > 0.5) between PC and PPP (see table). The values at 10 days of storage of approximately 11% of the native C3. Possible effects on stored platelets of C3 conversion in the surrounding plasma include: activation of platelets by C3a des-Arg (H. Marwijk Kooy, H.C. van Pollemy, H.A. van Riet, and T. Nachman; J. Exp. Med. 158:603, 1983) and deposition of complement factor C3b and C5b-9 (A. Salama and C. Mueller-Eckhardt, Transfusion 25:328, 1985).

In contrast, <0.01 ng/ml C5a was found in all samples tested, representing less than 0.2% conversion of C5a. Nephelometric assay of native C5 levels in PC samples showed a slight but significant difference by a paired t-test (p < 0.04) between fresh PC (mean±SD 1171 ng/ml ± 12.0, n=6) and PC stored for 10 days (mean±SD 108 ng/ml ± 9.7). No change in C5 levels was observed in stored PPP (mean±SD 156 ng/ml ± 10 ng/ml). Radiolabelled monoclonal antibodies to C3 fragments showed less than 600 molecules bound per platelet. This study demonstrates for the first time the extent of complement activation in stored platelet concentrates.

Repetitive transfections with platelets from randomly selected donors lead to HLA alloimmunization in about 50% of the patients. This is caused by lymphocytes that contaminate the platelet concentrates. Attempts to remove the leukocytes from the platelet concentrates by additional centrifugation steps lead to substantial loss of platelets.

We report here a new procedure for removal of almost all leukocytes with excellent platelet recoveries. Single donor concentrates are treated with 50 ng/ml prostacyclin in order to inactivate the platelets transiently. The concentrates are then passed through a cellulose acetate filter to remove the leukocytes. In 30 fresh concentrates this treatment reduced the contamination by leukocytes to less than 0.1 million per concentrate with a platelet recovery of 89 ± 7% (mean ± SD). In concentrates stored for 3 days the contamination was reduced to about 5 million per concentrate. Thirty filtered platelet concentrates, obtained from single donors by platelet apheresis using a Haemonetics U 50, were transfused to ten thrombocytopenic patients within 1 hour after filtration and were well tolerated. No signs of hypotension or other side effects were observed. The transfusions led to corrected count increments of (22.1 ± 1.1) x 10^9 per ml blood after one hour and normal survival thereafter. In four out of five patients these concentrates reduced the bleeding time.

We conclude, that transient inactivation of platelets by prostacyclin enables optimal removal of leukocytes and may help to reduce alloimmunization during frequent transfusions with platelet concentrates.

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**Tables and Figures**

**Table 1.** Comparison of in vivo and in vitro survival of platelets from normal donors.

<table>
<thead>
<tr>
<th>Day</th>
<th>PC (n=6)</th>
<th>PPP (n=6)</th>
</tr>
</thead>
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<tr>
<td>1</td>
<td>237</td>
<td>336</td>
</tr>
<tr>
<td>5</td>
<td>1038</td>
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<tr>
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</tr>
</tbody>
</table>

**Figure 1.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 2.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 3.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 4.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 5.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 6.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 7.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 8.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 9.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 10.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 11.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 12.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 13.** Comparison of in vivo and in vitro survival of platelets from normal donors.

**Figure 14.** Comparison of in vivo and in vitro survival of platelets from normal donors.