REPORT OF THE CYTOMEGALOVIRUS STEERING GROUP

March 2012
# Contents

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Introduction</td>
<td>3</td>
</tr>
<tr>
<td>Advice sought from SaBTO</td>
<td>4</td>
</tr>
<tr>
<td>Information on CMV biology and testing including PCR and serology</td>
<td>4</td>
</tr>
<tr>
<td>Filters and testing – efficacy and problems in the UK blood services</td>
<td>9</td>
</tr>
<tr>
<td>Specific patient groups</td>
<td>14</td>
</tr>
<tr>
<td>CMV Steering Group members</td>
<td>24</td>
</tr>
<tr>
<td>All references</td>
<td>25</td>
</tr>
</tbody>
</table>
INTRODUCTION

The SaBTO CMV (cytomegalovirus) Steering Group was set up to make recommendations on whether there is sufficient evidence to support the replacement of CMV seronagative cellular blood components (both red cells and platelets) with leucodepleted blood components. A meeting of the CMV Steering Group was held on 1 March 2011, chaired by Dr Mike Potter, and a follow up teleconference was held on 15 March 2011. The paper attached sets out the conclusions reached by the Steering Group for specific groups of patients.

The Steering Group recommends that:

1. CMV seronegative red cell and platelet components should be provided for intra-uterine transfusions and for neonates (ie up to 28 days post expected date of delivery), and therefore all small sized blood packs and other cellular blood components intended for neonates should be provided as CMV seronegative.

2. Granulocyte components should continue to be provided as CMV seronegative for CMV seronegative patients.

3. CMV seronagative blood components should be provided where possible for pregnant women, regardless of their CMV serostatus, who require repeat elective transfusions during the course of pregnancy (not labour and delivery). This mainly applies to patients with haemoglobinopathies who are managed in specialist centres. However CMV seronegative blood components are not expected to be generally available in all hospitals and therefore for emergency transfusions in pregnant women, leucodepleted components are recommended.

4. All blood components (other than granulocytes) in the UK now undergo leucodepletion, which provides a significant degree of CMV risk reduction. This measure is considered adequate risk reduction for all other patients requiring transfusion (haemopoetic stem cell transplant patients, organ transplant patients, and immune deficient patients, including those with HIV) without the requirement for CMV seronegative components in addition.

5. CMV PCR monitoring should be considered for all haemopoetic stem cell and solid organ transplant patients (even CMV negative donor/negative recipients) to allow early detection of any possible CMV infection (whether transfusion-transmitted or primary acquired infection).

6. Transfusion-transmitted CMV infections should be reported via the SHOT (Serious Hazards of Transfusion) and SABRE (Serious Adverse Blood Reactions & Events) systems.
ADVICE SOUGHT FROM SaBTO

The Steering Group sought the following advice from SaBTO:

1. Does SaBTO agree with each of the recommendations made by the CMV Steering Group?

2. If not, are there any amendments or alternative recommendations which SaBTO would make? If so, on what evidence or grounds does SaBTO make any amendment or alternative recommendation?

INFORMATION ON CMV BIOLOGY AND TESTING INCLUDING PCR AND SEROLOGY

Biology of CMV infection

Cytomegalovirus is a herpes virus, giving rise to chronic, persistent and for the most part asymptomatic infection in a majority of adults worldwide. Initial infection is most often subclinical, but occasionally presents as a febrile illness with mononucleolar syndrome in the normal host. Following primary infection, the host seroconverts, and CMV specific immunoglobulin G (IgG) persists lifelong together with cellular immune responses. A CMV seropositive individual is thus potentially infected and infectious for life.

Lifelong infection and reactivation facilitates transmission to intimate contacts. CMV may be transmitted horizontally in saliva and other body fluids, blood, haemopoietic stem cells and organ transplants. Primary infection occurs earlier in resource-poor countries and lower socio-economic groups. Seroprevalence in adults ranges from circa 40% (France, Germany) to 100% (Philippines, Uganda). In the UK circa 50-60% of UK adults are CMV seropositive, with an estimated seroconversion rate of circa 1% per annum. Seroconversion rates of 0.2-1.2% among blood donors have been reported from resource-rich countries, with NHS Blood and Transplant (NHSBT) estimating a rate of 0.28% in 2010 (personal communication S MacLennan).

Vertical transmission with postnatal infection through breastfeeding is an important route of transmission. Congenital infection also occurs, especially with primary maternal infection in pregnancy, and is not uncommon, with rates of 0.3% of live births in the UK and 1% of live births in the USA.

CMV manifests a broad cellular tropism in vivo, with the capacity for productive infection in a broad range of epithelial cells and in certain terminally-differentiated myeloid cells.

CMV persists in a latent state in CD34+ bone marrow progenitor cells and circulating monocytes (Taylor-Wiedeman, Sinclair & Sissons 2006, Sinclair 2008). Only a small proportion of circulating monocytes carry CMV, estimated
at between 1 in 10,000 and 1 in 100,000 peripheral blood mononuclear cells. These cells are non-permissive for manufacture of infectious CMV in vitro. Reactivation of productive infection requires expression of Immediate Early proteins occurring after terminal differentiation to macrophage/mature dendritic cells. Allogenic T cell stimulation of latently infected monocytes appears to fulfil the requirements, leading to new production of infectious virus. Other cells of the myeloid lineage do not support replicative CMV infection.

**Transmission of CMV in blood products**

Transmission of CMV present in blood products can give rise to primary infection in CMV naïve recipients (transfusion-transmitted CMV) or to reinfection in previously infected individuals.

The high risk of CMV transmission and disease in multiply-transfused CMV negative recipients was greatly reduced with the replacement of fresh blood products with stored blood products (Preiksaitis 1998) and with the provision of leucodepleted and/or CMV seronagative blood products (Drew 2007).

Although both approaches significantly reduce the risk of CMV transmission, neither is 100% effective, with an estimated risk of 1.5-3.0% per recipient in the setting of bone marrow transplant (Vamvakas 2005). Recent studies in blood donors help explain why.

**Why are there failures with either approach?**

CMV can be transmitted from blood donors with active (primary/reactivated) or latent infection. CMV may be present in circulating monocytes, or free in plasma as a result of primary infection or perhaps reactivation (see Figure 1).

**Figure 1**

Cytomegalovirus DNA in blood.
**Leucodepletion** diminishes the risk of CMV transmission but does not eliminate it.

Recent donor infection is likely to account for these failures. CMV DNA was repeatedly present in the plasma of 44% of newly seropositive donors with an estimated detection rate of 0.13% of all units due to primary CMV infection (Ziemann 2007). In a recent study of the natural course of CMV infection in blood donors, 13 individuals were followed prospectively. Of 148 new CMV seroconvertors among 17,982 donors, these 13 had laboratory evidence of ongoing primary infection. CMV DNA was detected in white cells and plasma for up to 269 days of follow-up and clearance was correlated with clearance of CMV IgM, and development of CMV glycoprotein B-specific IgG and high avidity IgG (Ziemann 2010, See Figure 2). The use of CMV DNA, IgM or avidity testing has thus been suggested as a means of identifying infectious donors.

![Figure 2](image)

**Figure 2**

Duration of primary CMV infection in newly seropositive donors with detectable plasma CMV viraemia.

Ziemann Vox Sanguinis (2010) 99, 24-33

**CMV seronagative donors** are at risk of acquiring infection between donations. Infectious virus may be present in the cellular and plasma fractions of donated blood during the estimated 6-8 week antibody window period of primary infection, prior to the seroconversion (Drew 2007, Ziemann 2007): see Figure 3. With CMV present in plasma fractions, infection might still be transmitted in leucodepleted blood donated by a “CMV negative” donor during the antibody window period.

By lessening the duration of the window period, NAT (Nucleic acid Amplification Technology) testing of donor blood could reduce this small risk.
While CMV viraemia precedes seroconversion and may persist for many months following primary CMV infection, CMV genomes are not detected in the plasma of donors with longstanding CMV infection (Drew 2007, Ziemann 2007). These observations have led to the controversial suggestion that leucodepleted blood products from donors who are CMV seropositive and known to have been seropositive for more than one year should be selected for vulnerable patient groups.

Alternative approaches include the use of NAT testing of whole blood and/or plasma to further increase the safety of leucodepleted blood products of known or unknown CMV serostatus.

Are CMV seronegative products more efficacious than leucoreduced blood components in preventing transfusion-acquired infection?

While there is a small risk of CMV transmission with either approach, it is not clear whether the use of CMV seronegative donors adds a significant advantage when blood is already leucodepleted. This situation is unlikely to change. As outlined by Vamvakas, 6,500 patients would need to be enrolled in a randomised controlled trial to have 80% power to detect a statistically significant (P = 0.05) difference (Vamvakas 2005).
Groups at risk of transfusion-transmitted CMV

The following patient groups are at risk of transfusion-transmitted CMV: CMV seronegative haematopoietic stem cell and solid organ transplant recipients (receiving CMV negative transplants), patients with malignant disease, low birth weight and/or premature neonates of CMV seronegative mothers, foetal transfusion recipients, foetuses of pregnant (CMV seronegative and seropositive) women.

CMV serological testing

As is the case for serological tests, with the exception of rubella IgG, there is no International Standard for comparison. In order to assess the validity of serological testing, Sester et al compared IgG results in two serological assays with the presence of CMV specific CD4+ cells in 388 individuals. Both assays largely agreed but two of 94 (2.1%) CMV IgG negatives were falsely negative on the basis of detectable cellular immunity (Sester 2003).

The Health Protection Agency evaluated six different CMV IgG/total antibody kits suitable for use in the blood service, and found all to have comparable sensitivities (98.51-100%). Specificities varied more widely, with the poorest (88.6%) being improved following notification to the manufacturers and subsequent assay adjustment. Taking the improvement into consideration, the new range of specificity was 93.38-100% (Curtis 2007).

CMV IgG avidity would allow identification and exclusion of recent seroconvertors. This test is now available in commercial robotic assays eg Abbott Architect, Diasorin Liaison.

CMV detection using Nucleic acid Amplification Technology (NAT)

Until now a variety of in-house and commercial qualitative and quantitative NAT assays have been developed and used to detect CMV DNA. CMV detection in plasma, mononuclear cells or whole blood has been reported in a variety of units with results usually reported as genome copy numbers with varying denominators including volume, cell count or µg DNA.

Variation in the sensitivity and specificity of PCR (polymerase chain reaction) assays has been associated with widely varying estimates of the frequency of detection of CMV DNA in blood donations (Larrson 1998, Roback 2001, Roback 2003).

International multisite comparisons of results using standard sample panels has shown marked inter-laboratory variation, and a lesser degree of intra-laboratory variation, especially for samples with lower viral loads approaching the limits of detection (Pang 2009, Wolff 2009). The need for a CMV DNA reference standard has finally been met by the National Institute for Biological Standards and Control with the development and provision of an International Standard for quantification of CMV viral load in clinical samples. As a result it will be
possible to report results in International Units, and make valid comparisons in the limits of sensitivity of different CMV NAT assays.

Use of CMV NAT detection to minimise transmission to high risk recipients is likely to generate a high proportion of initial false positives, as illustrated by a recent study using hepatitis B (HBV) DNA NAT to identify HBV infection in blood donations collected from HBV seronegative (s antigen and core antibody) donors during the HBV window period (Stramer 2011).

List of other evidence used (for example conversations, case studies etc)

See the sections on specific risk groups.

Any further work suggested for SaBTO

The Steering Group suggests the following further work:

1. Study of NAT testing of whole blood and plasma for IgM and IgG and IgG avidity, to find the most efficient method of further minimising risk in high risk groups for whom surveillance and pre-emptive therapy is not possible; and

2. Evaluation of the possible use of known longstanding CMV seropositive donors as a source of CMV-safer blood in situations where a lesser margin of error is still required.

FILTERS AND TESTING – EFFICACY AND PROBLEMS IN THE UK BLOOD SERVICES

Current provision for CMV risk reduction

Leucodepletion

Universal leucodepletion was implemented by all four UK Blood Services in 1999, primarily as a vCJD risk reduction measure. The UK specification for leucodepletion is $< 5 \times 10^6$ white cells per unit of platelets or red cells in 99% of components with 95% statistical confidence, and $< 1 \times 10^6$ white cells per unit in 90% of components.

The cut-off of $< 5 \times 10^6$ white cells per unit (3 log depletion) is generally accepted as the level which renders components “CMV safe” (Vamvakas 2005). This level was originally set because of experimental evidence relating to HLA (human leucocyte antigen) immunisation rather than to CMV transmission. Removal of CMV viral load has been demonstrated following 3-4 log white cell reduction of whole blood by filtration (Lipson 2001); and equivalent substantial reduction in CMV viral genome load was demonstrated following leucoreduction.
of platelets by apheresis, and filtration of platelets and red cells (Dumont 2007). However, a precise threshold level has not been identified.

CMV is thought to be latent in monocytes in the carrier (Taylor-Widerman 1991) and there is evidence to suggest that leucodepletion filters are particularly efficient at removal of monocytes from whole blood with $< 10^4$ monocytes per unit remaining post-filtration (Pennington 2001). A study investigating removal of CMV by leucodepletion filters using quantitative PCR demonstrated 2.8 log removal of CMV from ‘superinfected’ whole blood (Pennington 2001). The authors estimated that extrapolation of the results to the levels of CMV found in CMV carriers would suggest that no more than 0.01 to 0.1 viral copies per mL would remain in leucodepleted blood.

The efficacy of leucodepletion is monitored by testing a proportion of components using flow cytometry and monitoring using statistical process control. The chance of an issued component having a leucocyte count above the specification (corrected residual risk, CRR) can be calculated, and is a balance between the robustness and reliability of the leucodepletion process and the proportion of components tested. This varies between the UK Blood Services because of use of different pack and filter systems and different levels of testing. The table below provides recent data.

**Table 1**: Leucodepletion monitoring results from NHS Blood and Transplant, the Scottish National Blood Transfusion Service and the Welsh Blood Service, individual Service and combined data, Oct-Dec 2010 (no recent data available from Northern Ireland)

<table>
<thead>
<tr>
<th>Component</th>
<th>Blood Service</th>
<th>No. issued</th>
<th>No. tested</th>
<th>No. &gt;5x10^6/u</th>
<th>% &gt;5x10^6/u</th>
<th>No. &gt;100x10^6/u</th>
<th>CRR*</th>
</tr>
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<tbody>
<tr>
<td>Apheresis platelets</td>
<td>NHSBT</td>
<td>48374</td>
<td>10160</td>
<td>10</td>
<td>0.098</td>
<td>4</td>
<td>1:1286</td>
</tr>
<tr>
<td></td>
<td>SNBTS</td>
<td>5078</td>
<td>4833</td>
<td>4</td>
<td>0.083</td>
<td>0</td>
<td>1:25043</td>
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<tr>
<td></td>
<td>WBS</td>
<td>1684</td>
<td>237</td>
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<td>0.42</td>
<td>0</td>
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</tr>
<tr>
<td></td>
<td>Combined</td>
<td>55,136</td>
<td>15,230</td>
<td>15</td>
<td>0.098</td>
<td>4</td>
<td>1:1403</td>
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<tr>
<td>Pooled platelets</td>
<td>NHSBT</td>
<td>13480</td>
<td>1653</td>
<td>1</td>
<td>0.06</td>
<td>0</td>
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</tr>
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<td></td>
<td>SNBTS</td>
<td>2171</td>
<td>703</td>
<td>7</td>
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<td>0</td>
<td>1:149</td>
</tr>
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<td></td>
<td>WBS</td>
<td>557</td>
<td>52</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>&lt;1:52</td>
</tr>
<tr>
<td></td>
<td>Combined</td>
<td>16,208</td>
<td>2,408</td>
<td>8</td>
<td>0.33</td>
<td>0</td>
<td>1:354</td>
</tr>
<tr>
<td>Red cells</td>
<td>NHSBT</td>
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<td>4</td>
<td>0.033</td>
<td>0</td>
<td>1:3121</td>
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<td></td>
<td>SNBTS</td>
<td>59142</td>
<td>3425</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>&lt;1:3425</td>
</tr>
<tr>
<td></td>
<td>WBS</td>
<td>20302</td>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>&lt;1:1012</td>
</tr>
<tr>
<td></td>
<td>Combined</td>
<td>557,357</td>
<td>16,604</td>
<td>4</td>
<td>0.024</td>
<td>0</td>
<td>1:4278</td>
</tr>
</tbody>
</table>

* CRR = corrected residual risk of a component being issued with $> 5 \times 10^6$ leucocytes

Values $> 100 \times 10^6$/u indicate a failure of filtration or leucodepletion by apheresis. These are rare and can usually be picked up by visual inspection of
the filtration process, or in the case of apheresis platelets by the machine flagging a problem, in which case a leucocyte count will be performed on that component and the component will not be issued if it fails specification. The only failures at this level in the three month period Oct-Dec 2010 were apheresis platelets, and three of the four incidents were flagged by the apheresis machine. The risk of a high failure apheresis platelet component being missed and issued from one year’s UK data in 2010 is 1:7394.

Values > 5 but < 100 x 10⁶/u tend to be only just above the 5 x 10⁶/u level, particularly for red cells. Recent data from NHSBT show values of 5.0-8.6 x 10⁶ WBC/u for red cell components. Of six units of apheresis platelets, four were < 10x10⁶ WBC/u, and two were 30 x 10⁶ WBC/u.

**CMV antibody screening**

A proportion of donations are screened by the Blood Services for CMV antibody to provide a ‘CMV negative’ inventory of cellular components. Donations are selected from donors previously testing negative plus some new donors. The proportion of UK blood donors that are CMV antibody positive is about 25-40% depending on age group.

Antibody screening is performed using validated enzyme immunoassay (EIA) tests. These detect total CMV antibody (IgG and IgM) with a sensitivity of > 99.5% (equating to a ‘failure’ rate of < 1:200) and specificity between 98.1% and 99.3%.

CMV seronegative cellular components are provided to hospitals on request.

**Supply and cost of provision of CMV antibody negative components**

The number and proportion of CMV negative platelets and red cells has shown a gradual increase over the last five years and in 2009/10 amounted to 12% of red cells issued and 37% of platelets.

**Figure 4: CMV negative issues of red cells and platelets from NHSBT 2006-2010**
NHSBT charges £7.76 as a supplement for provision of CMV antibody negative red cells and platelets to cover the costs of screening and holding a separate inventory, leading to recovery of approximately £2.5 million per annum from hospitals of which £230,000 is for apheresis platelets and £2,270,000 for red cells (testing of which also provides buffy coats for pooled platelets, all four of which must be negative for the pool to be labelled as such).

More donations are screened than are issued as not all will be negative, and there needs to be sufficient to meet demand.

A decision to cease or reduce CMV testing will result in an immediate reduction in direct consumable costs as well as the potential, over time, to reduce other supply chain costs by reduction in wastage, reduced costs of platelet donor recruitment, and improved supply chain management and logistics. A 2% reduction in the rate of platelet expiry, for example, would save £0.3 million in supply chain costs.

**Advantages of a single inventory and accepting leucodepleted components as ‘CMV safe’**

Accepting leucodepleted components as CMV safe without requiring CMV seronegative components in addition has advantages for both blood banks and blood establishments.

**Inventory management**

Management of a single inventory would be less complex for both blood establishments and hospitals. The need to ensure local availability of CMV negative components on request means that Blood Services need to move components around the country between sites. In NHSBT it is estimated that approximately 50% of *ad hoc* deliveries to hospitals contain CMV seronegative platelets at a cost of approximately £95,000.

**Wastage**

It is estimated from data supplied by the Belgian Blood Service following the implementation of pathogen reduction of platelets (which inactivates CMV) that approximately 1.5% reduction in platelet wastage would be seen if the need to select for CMV negative was removed, at a saving of £0.22 million.

**Improved compliance with other safety initiatives**

The reduction in wastage and removal of the need for a second inventory would enable the target of 80% platelets by apheresis to be met more easily and consistently. This would also facilitate TRALI (transfusion related acute lung injury) prevention by making it easier to recruit enough male platelet donors, and would also avoid the costs of HLA antibody screening of female prospective platelet donors.
**Reduction in hospital blood bank workload**

The workload of hospital blood banks would be reduced as a result of the removal of the requirement to order or check CMV negative components, and stock management (including holding platelets as stock in the hospital) would be simplified.

**Reduction in clinical error**

Data from SHOT (Serious Hazards of Transfusion) from 2000 to 2010 showed that of 1,040 reports of ‘special requirements not met’, 83 were due to not selecting for CMV negative components when they were considered appropriate, and 65 due to not selecting for CMV negative plus irradiated components. None of these cases was reported as a CMV transmission.

**International practice**

Information was available from a European survey on provision of CMV safe components performed in 2010. Of 15 responders, 5 do not perform CMV serology testing at all but rely on leucodepletion. Of the 10 who did screen for CMV, all provided CMV negative components for intra-uterine transfusions, 8 provided CMV negative for neonatal transfusions and bone marrow transplant patients, and 6 provided CMV negative for pregnant females and immunosuppressed patients.

A similar survey conducted in the USA reported 65% of 183 institutions responding considered that CMV seronegative and leucodepleted components are equally effective in preventing transfusion-transmitted CMV (Smith 2010).

**UK Trusts’ requesting practices**

Clinicians in Oxford have recently agreed to accept leucodepleted components as CMV safe and not select for CMV seronegative components. Other regions are also considering their position in the light of this decision.

**List of other evidence used**

a. Leucodepletion efficacy – data collated and supplied by the UK Specialist Advisory Committee on Blood Components
b. Data on WBC levels in ‘leucodepletion failures’ - Mr Simon Procter, NHSBT
c. CMV screening – personal communication by Mr Steve Ramskill, NHSBT and paper from the Microbiological Diagnostics Assessment Service Evaluations and Standards Laboratory, Health Protection Agency Centre For Infections: “Evaluation of CMV kits suitable for use in UK Blood Service”
d. Cost of provision of CMV negative components – personal communication by Mr Vaughan Sydenham, NHSBT
e. Data on issues of CMV negative components – personal communication by Mr Michael Bowden, NHSBT
Advantages of single inventory – personal communications by Ms Lindsay Bissell, Ms Teresa Allen, SHOT

Data on clinical errors from SHOT – personal communication, Dr Sue Knowles

International practice – survey on provision of CMV safe components through the European Blood Alliance

Change of practice in Oxford – personal communication by Professor Mike Murphy.

SPECIFIC PATIENT GROUPS

Haemopoietic stem cell transplant patients – adults and paediatrics

For many years it has been standard practice that CMV seronegative recipients undergoing a transplant from a CMV seronegative donor are supported with CMV screened seronegative blood and platelet transfusions. Several studies have endorsed the rationale of this approach with rates of transfusion transmitted infection (TTI) less than 5% using screened/seronegative products versus 28%-57% with unscreened blood products (Bowden 1986, 1987; Miller 1991).

In 1995 a study based in Seattle (Bowden 1995) was the first and only large prospective randomised trial comparing the efficacy of CMV screened/seronegative blood and platelet support to the use of leucodepleted products using a bedside filter. This large study of 502 patients included adults and children undergoing both autologous and allogeneic transplantation for a variety of haematological malignancies. All recipients/donors were CMV seronegative as assessed pre-transplant. In this study CMV infection was defined by the presence of virus by culture or antigen detection from a clinical specimen. CMV disease definition required tissue biopsy evidence of CMV or the presence of this virus in bronchoalveolar lavage fluid together with compatible clinical/radiological changes. The primary end point was the incidence of infection or disease between day 21 and day 100 post-transplant. Earlier infections were excluded because they were considered unlikely to be due to transfusions.

In this study there were no significant differences between the probability of CMV infection (1.3% versus 2.4%, P=1.0) or CMV disease (0% versus 2.4%, P=1.0) between the seronegative and leucodepleted arms. There were no survival differences between the two groups. Therefore the primary conclusion from this study was that these two methods of mitigating transfusion-transmitted CMV were equivalent. However in the secondary analysis of all infections occurring from day 0 to day 100 post-transplant, although infection rates were similar, the probability of CMV disease in the leucodepleted arm was greater.
(2.4% versus 0% in the seronegative arm, P=0.03). However, the authors accepted that there were possible explanations for this; for example failure of the filter, and the higher percentage of repeat donations from single family donors used in the leucodepleted arm (the hypothesis being that repeat donations from a high level CMV secretor could have increased the risk of TTI). Also, this difference in disease rates was only evident when the whole transplant period (from 0 to 100 days) was considered, and it is possible that the early infections were due to false negative assessments of the recipients’ baseline serology status (with equivocal or discrepant serology test results in four of the five patients with early infections). Limitations of this study also related to the use of early prototypes of bedside filters (rather than the more efficient and reproducible methodology available today) and the lack of modern day CMV monitoring, eg by PCR, with early effective pre-emptive treatment. Other limitations relate to the inclusion of autologous transplant patients (in whom the incidence of CMV infection is very low) and the fact that in both arms errors occurred in allocation of intended blood products. The overall conclusion of this paper was that leucodepletion was an effective strategy to significantly reduce CMV TTI and screening/selection of seronegative products could be discontinued.

A second non-randomised study from the same centre but with different authors (Nichols 2003) looked at all CMV negative/negative transplants in two timed cohorts. In cohort 1 all blood products were CMV seronegative and/or filtered in the transfusion centre. In cohort 2, patients were also eligible to receive leucocyte-reduced platelets obtained by apheresis without additional filtration. Patients were closely monitored by a CMV antigen screen. The study found that the incidence of transfusion-transmitted CMV was higher in cohort 2 (4%) than cohort 1 (1.7%, P < 0.05). However, by multivariate analysis it was found that the use of filtered red blood cells from CMV positive donors was the primary predictor of transfusion-transmitted CMV (and not apheresis platelet products). As this study was not randomised it is possible that some other unidentified factor(s) may have changed between cohort 1 and 2 to explain the small difference in TTI. However, the use of pre-emptive therapy with Ganciclovir after detection of antigenaemia prevented all but one case of CMV disease prior to day 100 in the 807 patients, and reassuringly there were no CMV related deaths. This illustrates the importance of early CMV detection and effective treatment.

In a third but smaller and non-randomised study (Ljungman, 2002), there was no significant difference in infection or disease in CMV negative patients receiving screened plus filtered blood products versus leucodepleted products alone. In two cohorts of patients reported from Bristol (Foot 1998, Ronghe 2008), there was a zero incidence of CMV infection/disease in patients receiving CMV seronegative blood and platelets (Foot 1998), 110 allograft patients) or CMV seronegative red cells plus non-screened leucodepleted platelets (Ronghe 2008, 93 allograft patients). In these studies all patients and donors were seronegative and underwent regular CMV testing.
In conclusion, rates of CMV TTI have been very low with both leucodepletion and serology screening, and the two techniques are probably equivalent. With both approaches there is likely to be a low failure rate. CMV monitoring and early effective therapy may however be a successful strategy for mitigating against the potential clinical effects. Indeed the routine use of CMV quantitative PCR / pre-emptive therapy (eg with ganciclovir or valganciclovir) in the setting of stem cell transplantation has significantly reduced the mortality from CMV infection overall (even in transplants involving seropositive recipients and/or donors). The mortality from other viral infections (eg respiratory viruses), bacterial and fungal infections far exceeds that of CMV in current transplant practice.

In Seattle, the largest transplant centre in the world, seronegative screened products have been abandoned in favour of pre-storage leucodepletion (personal communication).

**SaBTO recommendations**

- CMV seronegative red cells and platelets may be replaced with leucodepleted blood components for adults and children post haemopoeitic stem cell transplantation, for all patient groups including seronegative donor/seronegative recipients.

- Patients requiring transfusions who may require a transplant in the future may also safely be transfused with leucodepleted products (eg seronegative leukaemia or thalassaemia patients).

- CMV PCR monitoring should be considered for all patients (even CMV negative/negative patients) to allow early detection of any possible CMV infection (whether transfusion-transmitted or otherwise acquired).

**Neonatal patients**

CMV is the commonest cause of congenital infection in the developed world, affecting 1–2% of infants worldwide, and has recently been reviewed in full by Luck and Sharland (2009). Up to 20% of babies who acquire congenital CMV die. CMV is estimated to cause up to 12% of all sensorineural hearing loss (SNHL) (Peckham 1987), and 10% of cerebral palsy. A high viral load is associated with a greater risk of SNHL, even if babies are clinically asymptomatic (Boppana 2005). There are large international differences in incidence, and the estimated frequency in the UK is 0.3–0.4% (Griffiths 1991). In the UK, just over half of all women presenting at antenatal clinics are seropositive for CMV (Tookey 1992).

Congenital acquisition of CMV occurs as a consequence of both recurrent and primary infection. Mothers are often asymptomatic with primary infection. Primary infection is more likely to cause symptomatic congenital CMV and long-
term sequelae than reactivation of infection. Primary infection may also increase the risk of abortion, stillbirth and fetal hydrops. Primary infection in the first trimester is more likely to result in neurological complications and SNHL (Pass 2006).

The risk of transplacental transmission during reactivated infection is around 1% (as opposed to as high as 40% during primary infection) but the high incidence of CMV seropositivity among pregnant women worldwide means that transplacental transmission during reactivated infection accounts for 30–50% of congenital infections, and there are well-documented cases of women having two affected infants.

Over 90% of infants with congenital CMV (culture and IgM positive) are asymptomatic in the neonatal period, but SNHL may be delayed and progressive (Pass, 2005).

In the minority who are symptomatic, severe multisystem disease may be present which is clinically similar to congenital rubella or toxoplasmosis. CMV hepatitis can lead to intrahepatic and extrahepatic bile duct destruction as well as haemochromatosis.

CMV infection of the fetal brain causes microcephaly.

The pattern of central nervous system damage probably varies with the timing of injury, so that lissencephaly is a feature of early injury, and polymicrogyria of slightly later injury. When the gyral pattern is normal, the injury has probably occurred in the third trimester of pregnancy. Hydrocephalus is also a feature of congenital CMV infection.

Eye involvement, with chorioretinitis, cataract and blindness, occurs in 10–20% of cases presenting in the neonatal period.

Pneumonitis may develop in the first few months after birth, even in infants who were initially asymptomatic.

The mortality from symptomatic neonatal CMV infection is between 10% and 30%, although much higher if the baby is premature. Thrombocytopenia and intra-uterine growth restriction are independent risk factors for SNHL (Rivera 2002). An abnormal CT or head ultrasound scan or abnormal BAER (brainstem auditory evoked response) also predict poor outcomes (Boppana 1997; Luck and Sharland, 2009).

It is unclear at what stage a baby born at term becomes less vulnerable to the effects of CMV infection. Babies most likely to be given blood products are those born preterm, or those who are sick. Concern has been expressed that such babies are likely to be at increased risk of the effects of CMV infection.

“Recent studies have not only defined factors important in the transmission of CMV, but also led to the suggestion of serious morbidity related to postnatal
acquisition. The burden of postnatal CMV disease and the risk-benefit of screening and prevention strategies are all still unclear.” (Luck and Sharland, 2009).

Consideration was given to estimating viral load using PCR in donors to babies in the neonatal period (first 28 days of life by definition for babies born at term; expected date of delivery + 28 days for those born preterm).

Postnatal infection is unlikely to result in the same consequences for the baby as infection acquired perinatally (Sharland and Griffiths, personal communication). It is probably unnecessary to continue to sero-screen blood for babies beyond the neonatal period (as defined above), who are still in their first year of life.

**Any further work suggested for SaBTO**

The Steering Group suggests further work to assess the additional actual (versus theoretical) risk of acquiring CMV perinatally if sero-testing is abandoned.

**SaBTO recommendations**

- There is discomfort amongst neonatologists about not being as sure as possible that blood products are CMV seronegative. There is insufficient evidence to support a change in current practice, though it is accepted that current practice may not necessarily protect a baby from early postnatal CMV infection secondary to transfusion, eg in cases where a previously seronegative donor becomes positive but has not yet seroconverted.

- Perinatally acquired CMV can have devastating consequences. Also PCR monitoring is not feasible in this patient group. Assurances will be needed that if a change in practice were to occur, it would not lead to any increase in risk of viral transmission.

**Pregnant patients**

**Summary of points answering whether there is sufficient evidence to support the replacement of CMV seronagative cellular blood components (for both red cells and platelets) with leucoreduced blood components for this patient group**

As many as 50% of pregnant women in the UK are CMV seronagative. Primary CMV infection in pregnancy is associated with a 40% risk of transmission to the fetus (Stagno 1986). Women who are already CMV seropositive can also transmit infection, in some cases following reinfection with a different strain of CMV (Boppana 2001).
Following primary maternal infection in pregnancy, 18% of neonates have clinical manifestations at birth (Fowler 1992). But some infants whose mothers had primary or recurrent CMV infection in pregnancy only present later, with 5-17% manifesting CMV sequelae including SNHL and developmental delay.

Appropriately-timed amniocentesis can rule out or confirm congenital CMV infection, but a positive result is not predictive of subsequent manifestations (Carlson 2010).

In a recently published systematic review of antenatal interventions for preventing transmission of CMV from mother to fetus and adverse outcomes in the congenitally-infected infant, the authors found no randomised controlled trial studies meeting the criteria for inclusion (McCarthy 2011). This contrasts with the successful prevention of CMV infection/disease by routine or pre-emptive prophylactic strategies in the post-transplant setting.

Emphasis has been placed on the importance of avoiding CMV infection or reinfection during pregnancy. In the USA, the American College of Obstetricians and Gynaecologists (ACOG) and Centers for Disease Control and Prevention (CDC) do not advise routine CMV screening in pregnancy but have advised on strategies to prevent CMV infection in women who are pregnant/planning pregnancy. These measures include education in hygiene around young children, avoiding sharing utensils and not kissing children of six years or younger on the mouth or cheek.

A single prospective study regarding transfusion-acquired CMV infection in pregnant women (Preiksatis 1998, summarised by Blajchman 2001) documented no seroconversions among 162 CMV seronagative pregnant women transfused with a mean of 2.7 units of red cells. Only 8 of the women were transfused prepartum however.

Pregnant women rarely require transfusion during pregnancy, the most common indications being in women involved in accidents or with pre-existing haemoglobinopathies. The lack of data on the risks of CMV reinfection among pregnant women with major thalassaemia had been raised in this context (Eleftheriou 1998).

List of other evidence used (for example conversations, case studies etc)

Professor Paul D Griffiths – advised use of CMV-negative products in all pregnant women
Dr Kate Langford – Consultant Obstetrician, Guy’s & St Thomas’ NHS Foundation Trust
Claire Harrison – Director of Haematology, Guy’s & St Thomas’ NHS Foundation Trust
**SaBTO recommendations**

- Despite leucodepletion, there is a small risk that CMV could be transmitted in blood products of recently infected donors, due to presence of plasma or the remaining white cell fraction. This risk applies in the 6-8 week antibody window period and continues for up to one year following seroconversion (Drew, Ziemann 2007, 2010). There is a high risk of congenital CMV infection following primary maternal infection, and of immediate or later sequelae in the fetus or neonate.

- Given the lack of successful interventions, it would seem prudent to continue to minimise this risk by the use of CMV negative blood products in addition to leucodepletion in pregnant women receiving elective transfusions. This only applies to transfusion during pregnancy, not for blood products required in association with labour and delivery. As CMV seropositive women are at risk of reinfection and vertical transmission of the newly-acquired CMV strain, CMV negative blood products should be requested for all elective transfusions during pregnancy, regardless of maternal CMV serostatus. This will avoid the need to determine maternal CMV status.

- If in an emergency situation it is not possible to provide CMV negative blood products, leuco-depleted products of unknown serostatus may be used.

**Intra-uterine transfusions**

Please see the section on transfusion in pregnancy above.

**Summary of points answering whether there is sufficient evidence to support the replacement of CMV seronagative cellular blood components (for both red cells and platelets) with leucoreduced blood components for this patient group**

The Steering Group was unable to find any data.

**List of published papers used in coming to this recommendation**

Please see the papers quoted in the section on pregnancy.

**List of other evidence used (for example conversations, case studies etc)**

Dr Pippa Kyle – Consultant in Maternal and Fetal Medicine, Guy’s and St Thomas’ NHS Foundation Trust

Professor Mark Kilby, incoming President of the British Maternal and Fetal Medicine Society, was going to put an enquiry out to the membership about CMV serology testing. His immediate reaction was that leucodepleted products would be acceptable in the light of the difficulties in serology testing.
**SaBTO recommendation**

- CMV seronagative red cells and/or platelets should continue to be supplied for intra-uterine transfusions, for the same reasons as cited in the pregnancy section. These are high risk, vulnerable patients with no effective predictive or therapeutic strategies available in the event of transfusion-transmitted infection.

**HIV patients – adults and paediatrics**

*Summary of points answering whether there is sufficient evidence to support the replacement of CMV seronagative cellular blood components (for both red cells and platelets) with leucoreduced blood components for this patient group*

The Steering Group was unable to find any relevant literature.

Due to the modes of HIV transmission, virtually all individuals with HIV infection also have CMV infection. Historically, the only exceptions were haemophiliacs and other individuals with transfusion-acquired HIV infection.

With routine antenatal HIV testing in the UK, vertical transmission is now rare. One paediatrician commented that infants with HIV infection usually already have CMV infection; another, that this is principally an issue pre-diagnosis of HIV. Nowadays these children are very rarely transfused.

The mainstay of CMV infection management is control of HIV infection. Effective treatment is available for CMV disease.

*List of published papers used in coming to this recommendation*

None

*List of other evidence used (for example conversations, case studies etc)*

Dr Nicholas Larbalestier – Clinical Lead, Genito-urinary and HIV medicine, Guy’s and St Thomas’ NHS Foundation Trust
Professor Adam Finn – Professor of Paediatrics, University of Bristol (Jackie Cornish)
Professor Mike Sharland – Professor of Paediatric Infectious diseases, St George’s Hospital
Dr Andrew Gennery – Clinical Reader in Paediatric Immunology and Haematopoietic Stem Cell Transplantation, Newcastle University (John Dark)
**SaBTO recommendation**

- CMV seronagative red cells and/or platelets should be replaced with leucoreduced blood components for both adult and paediatric HIV patients. The Steering Group, after discussion with other experts, agreed that there was no evidence to support the use of CMV negative blood for HIV positive patients. It was noted that it was very rare for HIV positive adults to be CMV negative. It was recommended that these patients should receive leucoreduced blood.

**Immunodeficient patients**

**SaBTO recommendation**

- The Steering Group agreed that there was no evidence to support the use of CMV negative blood for immunodeficient patients. It was recommended that these patients should receive leucoreduced blood.

**Organ transplant patients**

**Adult patients**

Of all the solid organ transplants, the lung is the most seriously affected by CMV infection. This is a reflection of the high level of immunosuppression required and the vulnerability of the lung itself to the damaging effects of infection. The transplanted organ has a range of impairments to local defences. The lung can therefore be used as a bellweather for potential effects on other organs.

Concern centres entirely around donor-acquired infections. The viral load transmitted in the lung, rich in lymphoid tissue and a reservoir for monocytes, is significant. A recent international survey of management practices did not mention transfusion-acquired infection (Zuk 2010).

Roughly half adult donors are CMV positive, as are half the recipients. Some subsets, such as patients with cystic fibrosis, are nearly all CMV negative. Approximately 25% of recipients will be donor positive, recipient negative, and therefore at risk of primary, donor-acquired infection, at a time when immunosuppression is at its highest level. This used to be a major concern, but has almost disappeared in the era of effective viral prophylaxis (Mitsani 2010, Manuel 2009). The same applies even to children, traditionally the most vulnerable group (Danziger-Isakov 2009).

None of these recent reviews mentions transfusion-acquired infection. Further evidence that it is of little concern is the very low incidence of seroconversion in
negative recipients who receive organs from seronegative donors. They receive no prophylaxis, or even monitoring. The rate of seroconversion is regarded as significantly less than the 1% per year quoted for their non-transplant equivalents.

There are no published studies comparing leucodepletion with CMV screening for blood or blood products given to lung transplant recipients. Given the very low risks involved, it is unlikely that any study could differentiate transfusion-from community-acquired infection.

Informal discussions with microbiologists linked to lung transplant programmes in the UK reveal no objection to the proposed SaBTO recommendations. If this is true for the lung, it is very unlikely that any different position would be encountered for the other organ transplant groups.

**Paediatric Patients**

CMV has a relatively high incidence of infection in patients receiving solid organ transplants. It is the most frequent infectious complication after solid organ transplantation. The risk varies with the type of organ received; the highest to lowest risk are lung, small intestine, pancreas, kidney, liver and heart.

CMV positive organs are used in CMV negative patients due to their scarcity and the high risk of patients dying on the waiting list. Hence primary CMV infection is given to some recipients during solid organ transplantation.

There is limited evidence of transfusion-transmitted CMV in the solid organ transplant population.

**SaBTO recommendation**

- The Steering Group agreed that there was no evidence to support the use of CMV seronegative blood for transplant patients. It was recommended that transplant patients should receive leucoreduced blood.

- Individual units should consider whether or not a policy of CMV PCR monitoring for some groups of patients (even CMV negative/negative patients) should be introduced to allow early detection of any possible CMV infection (whether transfusion-transmitted or otherwise acquired).
CMV STEERING GROUP MEMBERS

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Dr Eithne MacMahon Consultant Virologist
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