Winter 2010 • Issue 32

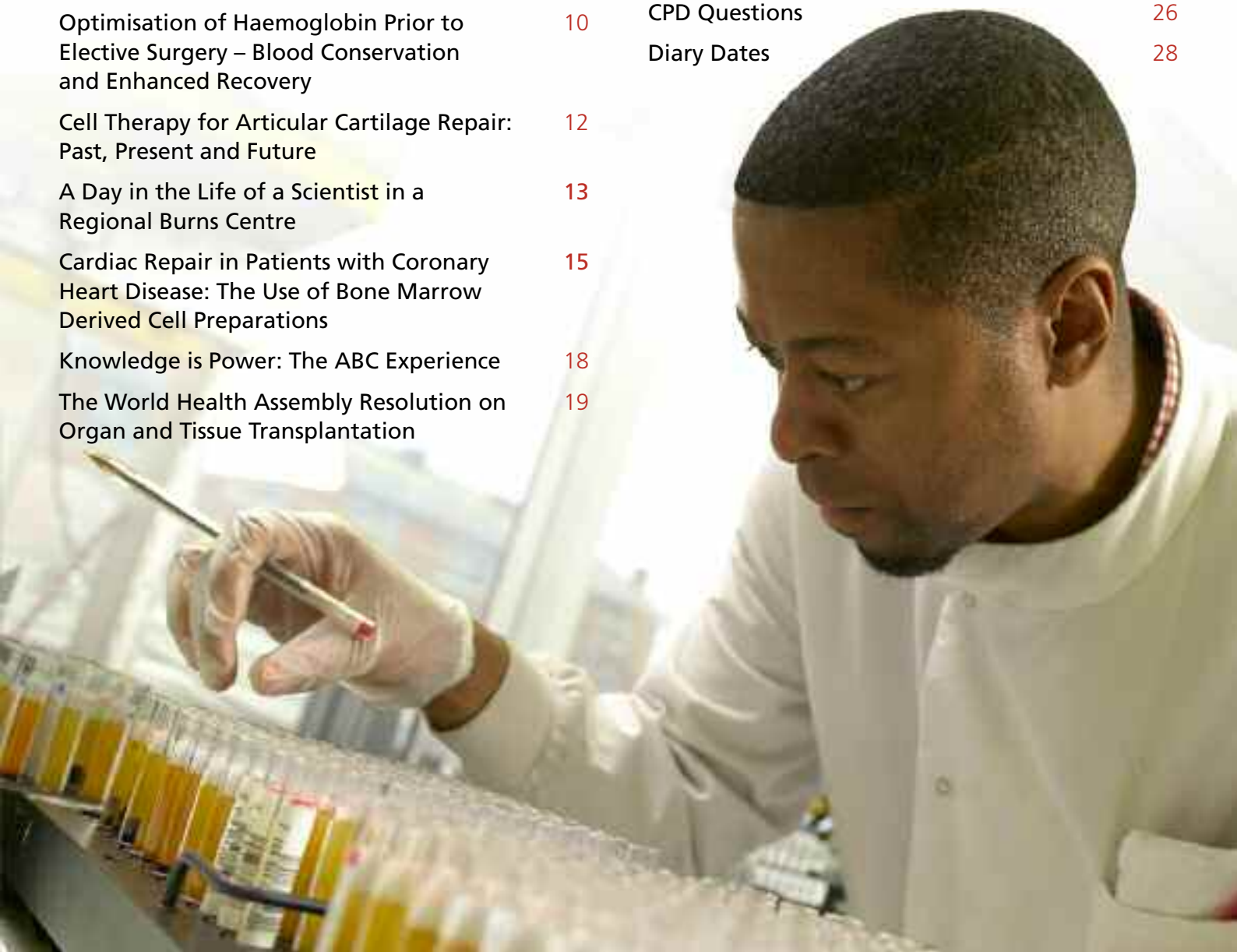
NHS Blood and Transplant

# Blood and Transplant Matters

Information for hospitals served by NHS Blood and Transplant

## Inside





# EDITORIAL

It is two and a half years since I joined the Editorial Board of *Blood and Transplant Matters*. When I was invited to write the 27th Editorial for the Spring 2009 issue, I commented then that ''Certainly blood matters! And tissues and organs also matter!" It was shortly after that the Editorial Board agreed to a name change for this journal to *Blood and Transplant Matters*. Each issue since then has increased in breadth and now there are regular articles on Tissues, Organs, Stem Cells, Advanced Therapies and Transplantation as well as Blood Component Collection, Production and Utilisation/Safety, Research and Audit.

In this Issue we have a clutch of important articles about blood component provision. Joyce Poole explains how patients with rare blood groups and antibodies to high incidence antigens and those transfusion dependent patients with high immunisation rates can be catered for when compatible blood is hard to find. Andrea Harris reminds us that guidelines for the administration of blood components were first developed ten years ago by the British Committee for Standards in Haematology (BCSH) and now, a decade later, they have been revised and updated. Her article highlights the key principles of blood transfusion and the principle changes between the 1999 and 2009 BCSH guidelines. Stephen Thomas discusses blood component development in NHSBT and how both laboratory studies and clinical collaboration are combined to show whether novel blood components are efficacious or not. Amber Raja, Lise Estcourt, and Simon Stanworth describe platelet transfusion thresholds, past and present prophylactic practice and why establishing thresholds is critical. Megan Rowley and Shubha Allard have written about how audits of blood transfusion in major surgery show huge variation for the same procedure in different organisations, how optimisation of haemoglobin in pre-operative patients can be achieved and how this may affect transfusion rates. I am pleased to say that we also feature one of Blood and Transplant Matters' favourite sections with a CPD questionnaire from Rob Webster.

As I retire from NHSBT and leave the Editorial Board, I am particularly pleased that there are also a number of articles about tissue bankers and tissue banking and more advanced cell therapies. We pay homage to two outstanding tissue bank innovators of their day. Professors Rudolf Klen (Czech Pioneer in Tissue Banking) and the distinguished scientist Professor Kazimierz Ostrowski (1921-2010 from Poland). These two gentlemen from Eastern Europe were pioneers in their field and modern day tissue banking owes much to them. They were both distinguished scientists who witnessed in the difficult circumstances of the Second World War. We are lucky to have an appreciation for Professor Ostrowski written by the accomplished tissue bankers Artur Kaminski and Janusz Komender from Poland and Professor Ján Koller from Bratislava has written an article about Professor Klen. Since their day, tissue banking has grown enormously. This issue also includes a description from Dr Debra Balderson, Head of Tissue Services, University Hospitals Birmingham NHS Foundation Trust of *''A Day in the Life of a Scientist in a Regional Burns Centre"* and articles on cell therapy for articular cartilage repair from Professor Cosimo de Bari and stem cells for treatment of heart disease by Dr Ranil de Silva.

Substances of Human Origin (SOHOs) raise many special ethical issues and the World Health Organisation (WHO) has maintained a global watching brief on the most important aspects. Jeremy Chapman from Australia, President of the Transplantation Society at the time of the World Health Assembly (WHA), has written an article for us about the WHA Resolution on organ and tissue transplantation. He urges us all to *''examine the WHO Guiding Principles carefully and thoughtfully and then to consider how well our own practices fit the global norms represented in this document."*

Jim MacPherson, Chief Executive Officer of America's Blood Centres (ABC) describes how the ABC views the dictum ''Knowledge is Power". ABC provides 10 million red blood cells, plus platelets and plasma to support nearly three million patients. He describes how the *ABC Newsletter* has become a weekly digest for tens of thousands of blood workers and hospital personnel around North America and beyond and its ethos shares much with our own Blood and Transplant Matters. Knowledge shared is knowledge gained and blood centres, transfusion laboratories and their professionals all gain from shared knowledge for the benefit of patients. The Editorial Board of Blood and Transplant Matters is proud that our journal is read by Jim MacPherson and that he has written for us in this issue.

The Editorial Board wish all our readers and contributors to issues over the last year a successful conclusion to 2010 and a happy and peaceful 2011.

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# Finding Compatible Blood for Patients with Difficult Antibodies

## *Introduction*

Patient's blood samples can be referred to the International Blood Group Reference Laboratory (IBGRL) red cell reference department for antibody identification, usually when compatible blood cannot be found. Referrals are made from within NHSBT and from other blood centres worldwide. In the context of this article, difficult antibodies are described in the two groups of patients that form the majority of our referrals; those with a rare blood group and whose plasma contains antibody to a high incidence antigen and transfusion dependent patients in whom immunisation rates are high.

Provision of blood for any patient with an alloantibody must take the clinical significance of the antibody into consideration. Clinical significance can usually be assessed reliably from serological reactions e.g. thermal range, mode of reactivity, reaction strength and from published data for an identified specificity. Antibodies vary widely in their clinical significance and there are guidelines which recommend when antigen-negative and cross match compatible blood should be selected for transfusion.

## *Patients with rare phenotype*

The serological investigation of patients with rare phenotype can be time consuming and complex. The complexity lies initially in the fact that all regular donor cells and routine panel cells are incompatible and antibody identification may be difficult. It is also important to establish whether underlying antibodies to more common specificities (e.g. K, $Fy^a$, $Jk^b$) are additionally present. If the antibody is identified and blood of the patient's rare phenotype is deemed to be a requirement but not available locally 'off the shelf', there are other options: The National Frozen Blood Bank in Liverpool; frozen blood banks in other countries e.g. The Netherlands, France, Spain, Japan; the National and International Panels of Donors of Rare Blood Type (which are compiled and maintained at IBGRL), possibly autologous donation or, on very rare occasions a family member, usually a sibling.

**Case Study 1:** Blood samples from a 74 year old British female patient, probable Rh phenotype $R1^wR1$, were referred with known anti-c+E+K but with an additional antibody reacting with all $R_1R_1$ K- cells. The patient was awaiting elective surgery for hip replacement. A clue to possible specificity was found when the $R1^wR1$ cell on a routine 10-cell panel was compatible. The patient and the compatible panel cell were subsequently shown to be homozygous for $C^w$ and thus have the rare $R_1^wR_1^w$ phenotype, which gives rise to lack of a high incidence Rh antigen, Rh51. Subsequent matching against other Rh:-51 cells, $Rh_{null}$, D- - and $R_1^wR_1^w$ (all extremely rare phenotypes), confirmed anti-Rh51. One unit of $R_1^wR_1^w$ K- blood was available from the UK National Rare Donor Panel and a further unit was obtained from Finland via the International Rare Donor Panel.

**Case Study 2:** Blood samples from a mother and her 15 day old baby, of African origin, were referred to us from Portugal. The baby had Haemolytic Disease of the Fetus and Newborn HDFN, Hb 5.6g/dl, and blood was required urgently. We identified the rare Rh e-related $hr^S$- phenotype in the mother and her serum contained anti-C+$hr^S$. Donors of $hr^S$- phenotype are very difficult to obtain in the UK and are most predominantly found in South Africa. A recently identified $Rh_{null}$ donor in Northern Ireland was called upon to donate a unit especially for the baby and the blood was shipped to Portugal. The baby's Hb rose to 11.8g/dl three days post transfusion and the baby continued to thrive.

## *Transfusion dependent patients*

Transfusion dependent patients e.g. with sickle cell disease (SCD), thalassaemia syndromes, severe aplastic anaemia, myelodysplastic syndromes and other congenital or acquired chronic anaemias can also cause problems with blood provision. These patients can be exposed to many allogeneic red cell antigens and alloimmunisation rates are high. Multiple antibody production can make antibody identification difficult. Additional problems arise in cases of SCD patients, usually of African origin, when they also have a rare blood group such as S-s-U-, Js(b-), $hr^S$-. Extended red cell phenotyping prior to initiating a transfusion regimen is recommended for these patients, to try and prevent alloimmunisation by providing the best matched blood and molecular genotyping can be used when transfused cells are present. Provision of blood for SCD patients can be especially difficult for several reasons: high incidence of alloimmunisation, high incidence of transfusion reactions and hyperhaemolysis even after transfusion of compatible blood, high incidence of autoantibody and mixtures of antibodies that change over a period of time. Closely antigen-matched and also ethnically matched donor units, commonly C-, E-, K-, Fy(a-) Jk(b-), S-, are often needed for these patients.

**Case Study 3:** Blood samples from a Senegalese SCD patient were referred to our laboratory from Spain. The patient had a Hb of 4.2g/dl and a strong antibody and that had caused a haemolytic transfusion reaction. Blood was requested urgently. Lengthy and time consuming

studies involving the use of rare cells, absorption/elution studies, skilled serology and acute observations revealed the presence of a complex mixture of two strong antibodies to high incidence antigens. Her cells had the rare Rh:32, -46 phenotype and anti-Rh46 and a Sc-related antibody were present. Both of these antibodies were considered to be highly clinically significant but no compatible donors were available. Fortuitously, blood from her sister was found to be compatible and one unit of her sister's blood was said to have ''brought the patient back to life".

### *Summary*

Antibodies of different blood group specificities vary widely in their clinical significance. The less common specificities, notably where all or the majority of cells are incompatible, can cause problems in antibody identification and a subsequent delay in patient care. If blood is not readily available locally there are panels of donors of rare phenotype, fresh or frozen, that may be called upon to provide blood for specified patients. Occasionally autologous units are used and on very rare occasions, a compatible sibling.

Transfusion-dependent patients may make multiple antibodies that change over a period of time and transfusing closely antigen-matched blood is recommended. Molecular genotyping is an option when transfused blood is already present and routine phenotyping is not possible. The cases described illustrate some of the complexities encountered in antibody investigation when a rare blood group is involved and the problems in obtaining compatible blood.

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**References**

Daniels G, Poole J, de Silva M, *et al*. (2002) The clinical significance of blood group antibodies. *Transfusion Medicine;* **12**:287-295.

Poole J and Daniels G. (2007) Blood group antibodies and their significance in transfusion Medicine. *Transfusion Medicine Reviews;* **21**:58-71

BCSH Blood Transfusion Task Force. (1996) Guidelines for pre-transfusion compatibility procedures in blood transfusion laboratories. *Transfusion Medicine;* **6**:273-283

NHSBT SPN/DDR/RC/020/03. The clinical significance of blood group alloantibodies and supply of blood for transfusion.

NHSBT SPN/DDR/RC/017/03. Provision of red cell transfusion support for transfusion dependent patients.

## BCSH Guidelines for the Administration of Blood Components

### *Introduction*

The administration of blood components is a critical task. Errors in the requesting, collection and administration of blood components lead to significant risks to patients. Since its launch in 1996, the Serious Hazards of Transfusion (SHOT) scheme has continually shown that 'wrong blood into patient' episodes are a frequently reported transfusion hazard. These wrong blood incidents are mainly due to human error leading to misidentification of the patient during blood sampling, blood component collection and delivery to the clinical area or blood administration and can lead to life-threatening haemolytic transfusion reactions and other significant morbidity.

Guidelines for the administration of blood components have been available from the British Committee for Standards in Haematology (BCSH) since 1999, and in 2009 these guidelines were reviewed and updated.

This article briefly discusses the key principles of safe blood administration and the main changes between the 1999 and 2009 BCSH guidelines.

The full BCSH guidelines for the administration of blood components (2009) are available from the BCSH website (**www.bcshguidelines.org**).

### *Key principles of blood transfusion practice*

The BCSH (2009) blood administration guidelines set out three key principles which underpin every stage of the blood administration process:

**1. Patient identification**

*Patient identification band (PIB):* In order to ensure positive patient identification, all patients receiving a blood transfusion must wear a PIB (or risk assessed equivalent) which contains the patients core identifiers (last name, first name, date of birth and unique patient identification number).

*Positive patient identification:* It is imperative that every patient has their identification checked and confirmed

correct at each stage of the transfusion process by asking them to state their full name and date of birth. This must match exactly the information on their PIB and any other associated paperwork required at that stage of the transfusion process.

**2. Documentation**

Full and complete documentation is required at every stage of the blood transfusion process to provide an assured and unambiguous audit trail. This should include the clinical reason for the transfusion, what was transfused and when and an indication of whether the transfusion achieved the desired effect. All paperwork relating to the patient must include the patient's core identifiers.

**3. Communication**

Clear and unambiguous communications between all involved in the transfusion process, including all clinical and laboratory staff and any other support staff, is essential. Care must be taken to avoid misinterpretation and transcription errors. Written, rather than verbal communications, may help to negate these errors.

## *Main changes between the 1999 and 2009 BCSH guidelines*

- There has been increasing experience of information technology (IT) solutions, currently based on electronically readable bar-codes or radiofrequency identification (RFID), to improve positive identification of patients, blood samples and blood components throughout the transfusion process. The 2009 guidelines advocate the use of IT to improve patient safety, but warns that all systems may have unintended risks as well as benefits. It is important to ensure that where such IT systems are used to enhance the blood transfusion process, that these systems are robust and are designed to meet the specific patient safety and identification requirements of transfusion medicine. Indeed, SHOT (2010) reported an error involving IT where a blood sample tube was incorrectly labelled (wrong blood in tube incident) which resulted in an ABO incompatible transfusion.
- The 1999 guidelines recommend the use of a patient identification number, but the 2009 BCSH guidelines discuss the use of a national unique identification number (such as the National Health Service (NHS) number in England and Wales, Community Health Index (CHI) number in Scotland, or Health and Social Care (HSC) number in Northern Ireland). The routine use of a national identification number should reduce the confusion caused by multiple hospital numbers and case records for the same patient.
- The NPSA Safer Practice Notice 14 (2006) 'Right patient, right blood' stipulates three yearly competency assessments for all staff involved in the blood transfusion process. The BSQR (SI 2005 No.50 as amended) also requires 'regular' competency assessment for the collection and distribution of blood components. In addition to recommending these competency requirements, the 2009 guidelines also emphasise the need for regular training. The 1999 guidelines did not specify how often training should be undertaken, but the 2009 guidelines now recommend that all staff involved in the blood transfusion process in the clinical area should receive at least one update training episode in-between the three yearly NPSA required competency assessments (thus an individual receives training and/or a competency assessment at least every two years).
- The 1999 guidelines stated that the prescription of blood components is the responsibility of a doctor. However, blood components are excluded from the current legal definition of medicinal products. This means that there are no legal barriers to other appropriately trained competent registered practitioners ordering, authorising and administering blood. Therefore, the 2009 guidelines emphasise the need for organisations to develop clear policies to extend the authorisation of administration of blood components safely and conveniently to other appropriately trained and competent practitioners, for example, authorised named nurses.
- The 2009 guideline recommends that minimum patient observations during transfusion episodes should now include baseline measurement of respiratory rate. This is in addition to the previous recommendation of temperature, pulse and blood pressure. These baseline measurements should be taken no more than 60 minutes before commencing the transfusion.

Observations at 15 minutes now also include blood pressure in addition to temperature and pulse. Regular visual observation throughout the transfusion is re-emphasised.

Post transfusion observations (temperature, pulse and blood pressure) should be taken no more than 60 minutes after the end of the transfusion episode.

It is now recognised that adverse reactions may manifest many hours after the transfusion is completed. The 2009 guidelines recommend that patients, such as day cases, discharged within 24 hours of transfusion are issued with a 'contact card' giving 24-hour access to clinical advice.

- The 2009 guideline no longer includes the management of transfusion reactions. These will be the subject of a separate BCSH guideline due later this year.

### *Conclusion*

The systems and processes involved in the transfusion pathway are very complex. Organisations should focus on simplifying procedures and concentrate on key steps, especially positive patient identification.

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**References**

Blood Safety and Quality Regulations (BSQR) 2005: Statutory Instrument 2005/50 (ISBN 0110990412) www.opsi.gov.uk/si/si2005/20050050.htm

British Committee for Standards in Haematology Blood Transfusion Taskforce (1999) The administration of blood and blood components and the management of the transfused patient. *Transfusion Medicine,* **9**, 227-238 http://www.bcshguidelines.com/documents/blood_administration_1999.pdf

British Committee for Standards in Haematology Blood Transfusion Taskforce (2009) Guideline on the administration of blood components. http://www.bcshguidelines.com/documents/Admin_blood_components_bcsh_05012010.pdf

National Patient Safety Agency (2006) *Safer Practice Notice 14: Right patient, right blood*. October 2006. NPSA/2006/14. http://www.nrls.npsa.nhs.uk/resources/?entryid45=59805

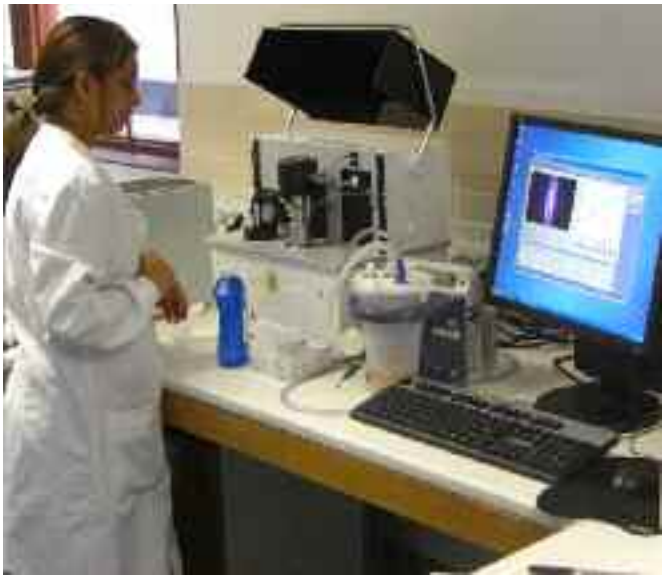
Serious Hazards of Transfusion (SHOT) scheme (1996 - 2010) *SHOT Annual reports 1996 - 2009*. SHOT office. Manchester. http://www.shotuk.org/home

## Component Development in NHSBT

NHSBT is constantly seeking to improve the safety, sufficiency and efficacy of the blood components supplied. Any procedural changes that may have an impact on the quality of the components must be fully validated before they may be implemented in a supply chain that handles millions of units every year.

The Component Development (CD) function is NHSBT's link between research, development, and routine operational use of new technologies or practices for blood component production. A good example of our work is the development of a new Pooled Granulocyte component, which may replace the current practise of transfusion of buffy coats. This component was developed in the laboratory, has just completed a clinical safety trial and will soon appear as a recognised component in the Guidelines for Blood Transfusion Services in the UK (the Red Book).

The majority of CD staff work in the Component Development Laboratory (CDL) in the Brentwood centre, a national facility equipped to prepare and analyse novel components in order to determine if they are fit for purpose. The work performed by CDL is guided by Chapter 9 of the Red Book, which makes clear that significant changes to any part of the component production process require in-depth laboratory analysis before progression to operational studies. As well as laboratory studies, we also collaborate with clinical colleagues based at NHSBT's Cambridge centre, to investigate whether novel components are safe and effective or not.



Decisions on what work CD should perform, and recommendations based on the outcome of the studies, are made through the Component Strategy Group, which also has representatives from Patient Services, Manufacturing, Clinical, Quality Assurance and Hospital Liaison. When it is relevant to do so, CD staff are encouraged to present their work at conferences and to publish their findings in peer-reviewed journals. This ensures that other blood services do not waste resources repeating similar studies, and also serves as a quality control on the standard of work performed.

Below are a few examples of recent CD projects that have had an impact on the blood components NHSBT supplies.

## *Prion reduction of red cell concentrates*

The DH advisory committee on the Safety of Blood, Tissues and Organs (SaBTO) recently recommended that red cell concentrates for patients born after 1 January 1996 and haemoglobinopathy patients should be processed using a prion reduction filter. Aside from the complex issue of proving the efficacy of the available filter (the Macopharma P-Capt), it was important to study whether there were any unexpected effects on the red cell concentrates (RCC) that were being subjected to this additional process. This project involved collaboration with colleagues in Processing and Red Cell Immunohaematology departments, and together we found that there were no significant effects on the quality of the RCC, apart from the loss of 5 – 7g of haemoglobin due to retention of some cells in the filter.

The work on prion reduced components is continuing, as the P-Capt filter also removes some clotting factors from the residual plasma left in the RCC. This has presented an opportunity to develop a simple quality monitoring assay to check that units have been correctly filtered. However, the removal of clotting factors also presents a challenge for RCC for neonatal exchange transfusion, which need to contain sufficient coagulation factors otherwise FFP may have to be transfused simultaneously. Development work is ongoing, to optimise the manufacture of a prion reduced red cell component for exchange transfusion, which can be made by re-suspending prion-filtered red cells with imported plasma.

## *Operational flexibility and efficiency*

For many years, NHSBT manufactured plasma and platelet components (as well as RCC) on the day of blood collection, and refrigerated any unprocessed blood for manufacture into RCC (only) on the following day. However, this process presented challenges in maintaining adequate stocks of all components, and the selection of male-only plasma to reduce the risk of TRALI. The development of NHSBT's large centre at Filton also required a more flexible approach to blood processing times. CDL therefore investigated, and validated, the European practice of 'ambient hold' of whole blood for up to 24 hours before component manufacture.

RCC for exchange transfusion that have not been used within their five day shelf life could be used for transfusion to adults, but this is not in keeping with current objectives to reduce patient exposure to donor plasma, to reduce the risk of vCJD or TRALI. CDL therefore conducted a study that showed these units could be remanufactured into standard RCC in additive solution, with no detrimental effects, or that leucodepleted whole blood could be held for five days before manufacturing, on demand, into either RCC for exchange transfusion or red cells in additive for other patients. This simple study has reduced unnecessary wastage of RCC.

## *Storage temperature deviations*

We have recently performed some studies on the effects of short term deviations in the normal storage temperature of RCC and Fresh Frozen Plasma (FFP). Although these studies have not yet been published, we are hopeful that when the work is complete, it will either allow revision of guidelines in this area or provide evidence for use in risk assessments when unplanned deviations do occur. These can occur either in the blood centre or hospital blood banks as a result of fridge or freezer malfunctions. Our hope is that this will also reduce unnecessary wastage of blood components.

These are just a few examples of how a relatively small in-house team of experts contribute to the safety and quality of the blood components that are provided every day by our front line staff in the supply chain. We report our findings through internal reports, published papers and conference presentations, and welcome feedback or suggestions for future work.

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### References

Bashir S, Stanworth S, Massey E *et al* (2008) Neutrophil function is preserved in a pooled granulocyte component prepared from whole blood donations. *Br J Haematol*, **140**, 701-11.

Joint UKBTS/NIBSC Professional Advisory Committee. Guidelines for the Blood Transfusion Services in the United Kingdom. **7 ed** (2005). London: The Stationery Office, 2005.

Wiltshire M, Thomas S, Scott J *et al* (2010) Prion reduction of red blood cells: impact on component quality. *Transfusion*, **50**(5), 970-9.

Wilsher CK, Garwood M, Turner C *et al* (2008) The effect of storing whole blood at 22°C for up to 24 hours with and without rapid cooling on the quality of red cell concentrates and fresh frozen plasma. *Transfusion*, **48**, (11) 2338-47.

Wiltshire M, Cardigan R, Thomas S (2010) Manufacture of Red Cells in Additive Solution from Whole Blood Refrigerated for Five Days or Remanufactured from Red Cells Stored in Plasma. *Transfus Med*, **20** (6), 383-91.

# Platelet Transfusion Thresholds

## Introduction

Platelet transfusions are given to a range of patient groups including cancer, trauma and surgery patients. The majority of platelet transfusions, in the UK, are used to prevent bleeding from occurring (prophylactic) rather than to treat active bleeding (therapeutic). A patient's platelet count is usually the deciding factor when considering whether a patient requires a prophylactic platelet transfusion. With the platelet count having such a pivotal role in the decision to transfuse, it is important to establish a clear definition of the platelet count where the benefits of a platelet transfusion outweigh the risks. This will guide the appropriate use of platelet transfusions as platelet concentrates carry a low but measurable risk of adverse effects, as well as being a limited and expensive resource.

This article will give a brief overview of past and present prophylactic platelet transfusion thresholds and their importance.

## Adults and children

### *A Brief History*

Until recently, the most widely quoted transfusion threshold for adults and children has been 20 x $10^9$/L, which was based largely on the 1962 paper by Gaydos *et al*. This paper reported that gross haemorrhage rarely occurred at platelet counts above 20 x $10^9$/L[2]. However, the paper specifically stated that "no 'threshold' platelet level was observed" during this trial. Despite this, the use of 20 x $10^9$/L as the prophylactic platelet transfusion threshold became widespread in the 1970s and 1980s, causing an increased demand for platelet concentrates. Because of this increased demand for platelets and concerns regarding unnecessary patient exposure to the risks of transfusion, clinicians started to re-examine the use of the 20 x $10^9$/L platelet threshold in the 1990s.

### *Current Practice*

There have been several studies re-examining the 20 x $10^9$/L platelet transfusion threshold that have all stated that this figure could be safely lowered to 10 x $10^9$/L. The most cited of these studies is the Platelet Transfusion Trigger Trial (PTTT) by Rebulla *et al* in 1997. This multi-centred randomised controlled trial (RCT) compared a transfusion threshold of 20 x $10^9$/L (liberal group) with a transfusion threshold of 10 x $10^9$/L (restrictive group). The results of the study concluded that there was no significant difference between the two groups in the number of major bleeding episodes. This study, together with others, helped to form the current evidence-based consensus for platelet transfusions in adults and children.

**British Committee for Standards in Haematology (BCSH) 2003 Guidelines for the Use of Platelet Transfusions.**

Evidence-based recommendation – *"patients should receive a platelet transfusion when their platelet counts are below 10 x $10^9$/L, unless suffering from a condition that necessitates transfusion at a higher platelet count (sepsis, fever etc)."*

## Neonates

### *A Brief History*

Although consensus has been reached for platelet transfusion thresholds in adults and children, wide variation still exists in platelet transfusion thresholds for neonates. The already established transfusion thresholds for adults and children cannot simply be applied to neonates due to substantial developmental differences in platelet function and haemostasis between these patient groups.

Over the last decade or so, several studies have revealed the extent of variation that still exists worldwide in the use of neonatal platelet transfusions. One such study is the web-based survey by Josephson *et al*. 2009, which revealed significant diversity in transfusion thresholds. For instance, for stable, pre-term neonates (27 weeks gestation) transfusion thresholds ranged from 10 x $10^9$/L to 150 x $10^9$/L[3]. The survey concluded that the underlying cause of high diversity in platelet transfusion thresholds reflected the lack of scientific evidence available to guide clinical practice. Therefore, prospective randomised clinical trials to generate evidence-based neonatal platelet transfusion guidelines are required.

### *Current Practice*

Although more restrictive transfusion policies are recommended in many guidelines there is a tendency towards more liberal use by neonatologists. The clinical uncertainty and variation in neonatal platelet transfusion practice, as well as the lack of evidence to support liberal transfusion practices has driven the need for clear neonatal transfusion guidelines.

A study that aims to define safe platelet transfusion support for neonates (Platelets for Neonatal Transfusion (PlaNeT) study 2) is due to start later this year. This trial will compare two platelet transfusion thresholds (25 x $10^9$/L and 50 x $10^9$/L), and establish whether a lower platelet transfusion threshold is as safe as a higher threshold. The trial follows on from a prospective multicentre observational study of platelet transfusions practices in neonates, which, like Josephson and

colleagues, found that policies and protocols for neonatal platelet transfusions vary widely between clinicians and institutions. The findings from this clinical trial could be instrumental in establishing evidence-based guidelines for neonatal platelet transfusion thresholds.

BCSH 2004 Transfusion Guidelines for Neonates and Older Children

Expert opinion based recommendation – *"term infants are unlikely to bleed if the platelet count is maintained above 20 x $10^9$/L, but in small, preterm babies a higher threshold is generally recommended".*

## Conclusion

Positive efforts have been made to provide an evidence-base for prophylactic platelet transfusion thresholds in adults and children, and in neonates, with consensus being reached for the former. However, there continues to be debate on whether prophylactic platelet transfusions should be used at all. This debate has arisen amid concern that many clinically stable patients receive prophylactic platelet transfusions unnecessarily, exposing them to transfusion associated risks without conferring significant clinical benefit (i.e. decreasing their risk of bleeding). Several clinical trials which are ongoing will address this concern. One of these is a multi-national RCT (Trial of Platelet Prophylaxis (TOPPS)) in adults that aims to determine whether a no prophylactic platelet transfusion policy (i.e. platelet transfusions given only to patients who are actively bleeding) is as clinically effective and safe as the prophylactic platelet transfusion policy in current use. It is hoped that the results of this trial (expected in 2012), and others, will make a contribution towards the development of rational, evidence-based, and cost-effectiveness platelet transfusion guidelines.

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**References**

Rebulla, P., Finazzi, G., Marangoni, F., Avvisati, G., Gugliotta, L., Tognoni, G *et al* (1997) The threshold for prophylactic platelet transfusions in adults with acute myeloid leukemia. *The New England Journal of Medicine*, **337**, 1870-1875.

Gaydos, L.A., Freireich, E.J. & Mantel, N. (1962) The quantitative relation between platelet count and hemorrhage in patients with acute leukaemia. *The New England Journal of Medicine*, **266**, 905-909.

Josephson, C.D., Su, L.L., Christensen, R.D., Hillyer, C.D., Castillejo, M.I., Emory, M.R *et al* (2009) Platelet Transfusion Practices Among Neonatologists in the United States and Canada: Results of a Survey. *Pediatrics*, **123**, 278-285.

Stanworth, S.J., Clarke, P., Watts, T., Ballard, S., Choo, L., Morris, T *et al* (2009) Prospective, observational study of outcomes in neonates with severe thrombocytopenia. *Pediatrics*, **124**, e826-e834.

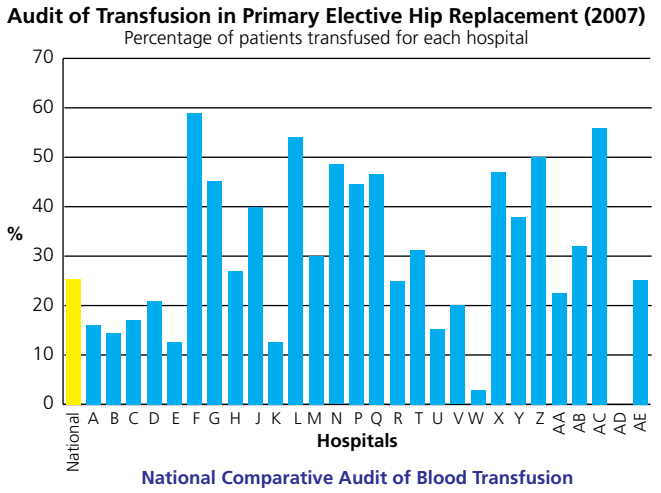
PlaNet-2 study protocol – further details from corresponding author (Amber Raja)

Stanworth, S.J., Dyer, C., Choo, L., Bakrania, L., Copplestone, A., Llewelyn, C *et al* (2010) Do All Patients With Hematologic Malignancies and Severe Thrombocytopenia Need Prophylactic Platelet Transfusions?: Background, Rationale, and Design of a Clinical Trial (Trial of Platelet Prophylaxis) to Assess the Effectiveness of Prophylactic Platelet Transfusions. *Transfusion Medicine Reviews*, **24**, 163-171.

# Optimisation of Haemoglobin Prior to Elective Surgery – Blood Conservation and Enhanced Recovery

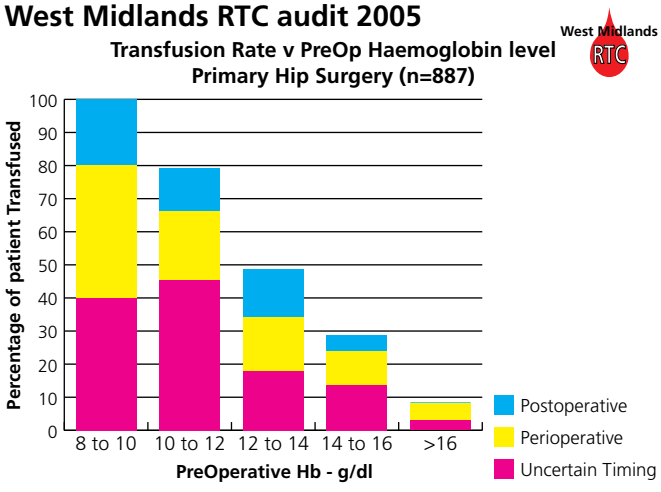
## *Background*

Audits of blood use in major surgery have shown considerable variation in transfusion rates for the same procedure undertaken in different organisations as demonstrated by the National Comparative Audit of Blood Transfusion (NCABT) in primary elective hip surgery.




National and regional audits also demonstrate that anaemic patients undergoing major surgery are likely to be transfused at some stage during their admission. In the West Midlands Regional Transfusion Committee (RTC) audit of blood use in orthopaedic surgery, patients with Hb 10-12g/dL had an 80% chance of being transfused; for Hb below 10g/dL, 100% were transfused. Although successful blood conservation measures have resulted in lower surgical transfusion rates in some centres, managing anaemia as a strategy is often overlooked. In the NCABT hip audit anaemia was common; overall 15% had a pre-operative Hb <12g/dL.

### West Midlands RTC audit 2005




The *Better Blood Transfusion* Health Service Circulars in 2002 and 2007 recommended that anaemia should be diagnosed and corrected in advance of surgery and it is well recognised by surgeons, anaesthetists and pre-operative assessment services that patient outcomes are improved if pre-operative health is optimised. The Association of Anaesthetists of Great Britain and Ireland include this advice in their guidance on Red Cell Transfusions (2008) and the British Orthopaedic Association, in their 2005 guidance on Blood Conservation in Elective Orthopaedic Surgery, state the expectation that anaemia should already have been recognised and treated by the time patients are seen in the preoperative assessment clinic.

## *Optimisation of Hb in Orthopaedic Surgery*

In 2009, a working party of the NHSBT Appropriate Use of Blood Group (AUBG) working with the West Midlands RTC Audit Group set out to find ways of improving preoperative management of anaemia by implementing and promoting existing evidence based guidance, concentrating initially on elective orthopaedic surgery. The basis for this work was the 2007 West Midlands RTC guidelines for the Management of Anaemia in Preoperative Assessment Clinics. This provides a template outlining when, where and how anaemia in the preoperative setting should be managed. At about this time, the Department of Health introduced the 18-week referral-to-treatment-time target with the result that stages on the surgical pathway needed to be shorter, allowing less time for optimising preoperative health. The AUBG made contact with the DH team working to improve compliance with this target for orthopaedic surgical pathways to raise the awareness of anaemia as an important area for preoperative management. As a result of this collaboration, the advice to optimise haemoglobin (Hb) PRIOR to referral for surgery has been included in recommendations to commissioners when placing contracts for surgery. The AUBG has also been actively involved in the Enhanced Recovery Partnership Programme, as outlined below, and the awareness of the importance of identifying and treating anaemia has been disseminated to a wider group including GPs, commissioners, managers, pre-operative assessment services and anaesthetists as well as surgeons and haematologists.

The recommended approach to optimisation of Hb prior to surgery that has been promoted is simple and clear;

**1. Patients referred for major surgery should be screened for anaemia as early as possible, and ideally in primary care, before the referral is made.** The GP is best placed to prioritise the patient's need for general health optimisation against the need for surgery. Although surgical referral may be delayed, this can be managed positively. If anaemia is not detected until the pre-admission visit, it may be too late to optimise Hb and

transfusion at some stage during the perioperative period is more likely. Alternatively, the decision may be made to cancel surgery and refer back to the GP for investigation which has a negative impact on the patient's experience.

**2. A pre-operative patient should be considered to be anaemic when the Hb is less than 12g/dL in a woman and less than 13g/dL in a man (WHO).** All too often, the arbitrary Hb taken as a trigger to investigate anaemia is <10g/dL, which is too low.

**3. Most anaemia is iron deficiency, or has an element of iron deficiency, and oral or intravenous iron should be considered.** Once identified, the sooner anaemia is treated, the more cost effective it is.

Improving iron in the diet is a good general pre-operative principle and NHSBT provide a useful patient information leaflet that can be used in GP surgeries and surgical clinics. Oral iron is inexpensive but may have gastrointestinal side effects. It can be prescribed by the GP or patients can be told to self medicate. Intravenous iron is more effective than oral iron, particularly in patients with iron utilisation block, and works more quickly. This is not widely used because of the resources required and to administer this treatment is problematic. Any iron treatment should be accompanied by a plan to investigate the cause as well as to assess the efficacy.

## *Enhanced Recovery*

The Department of Health, working with primary care and surgical teams, has developed enhanced recovery (ER) pathways. This is a multi-faceted where improved quality of care throughout the elective surgical pathway drives down the length of stay, improves surgical outcomes and provides an enhanced patient experience. Part of this approach is to physically and physiologically optimise patients preoperatively and psychologically prepare them for their forthcoming procedure. The ER process is summarised in a DH booklet and this includes input from the AUBG initiative on Hb optimisation, along the lines outlined above. Although uptake from the primary care community has been slow, GPs have been attending the ER launch conferences and work continues to raise the profile of ER, including Hb optimisation, within primary care teams.

## *Summary*

We know that managing anaemia preoperatively is good for patients and reduces transfusion rates.

The profile of this approach has been raised by the work of the Appropriate Use of Blood Group and the West Midland RTC with the expectation that it will become embedded in preoperative management and should, more often, be undertaken prior to surgical referral.

The West Midlands RTC guidance contains helpful practical advice and will be updated by a BCSH guideline in due course.

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### References

Blest, A, Baker, C, Taylor, C, Brothwood, D, Bramhall, J. November 2007 West Midlands Regional Transfusion Committee (RTC) guidelines for the management of anaemia in pre-operative assessment clinics. Access at: http://www.transfusionguidelines.org.uk/docs/pdfs/rtc-wmids_edu_anaemia_guide_preop_07_11.pdf

Delivering Enhanced recovery. Helping patients to get better sooner after surgery. Enhanced Recovery Partnership Programme March 2010 Gateway reference 13949. Access at: http://www.dh.gov.uk/prod_consum_dh/groups/dh_digitalassets/documents/digitalasset/dh_119382.pdf

# Cell Therapy for Articular Cartilage Repair: Past, Present and Future

## *Autologous Chondrocyte Implantation (ACI): gold standard of cell therapy for cartilage repair*

Post-traumatic osteoarthritis (OA) represents 13% of knee OA and 73% of ankle OA. Symptomatic chronic full-thickness defects of the knee joint surface require surgical treatment for both symptom relief and to prevent possible evolution towards OA.

In 1994 Brittberg and colleagues reported treatment of focal joint surface defects in humans by implantation of autologous, culture-expanded articular chondrocytes underneath a periosteal flap. Chondrocytes were obtained with a cartilage biopsy from a healthy and minor-load bearing area of the same joint surface. This pilot study showed symptomatic relief in 14 out of 16 patients with lesions of the femoral condyle at two years follow up.

So far, ACI has been carried out in more than 30,000 patients worldwide. Despite the lack of conclusive evidence supporting its clinical superiority to other far less expensive treatments such as microfracture (a surgical procedure to repair articular cartilage by creating tiny fractures in the underlying bone so that new cartilage develops from a so-called 'super-clot'), ACI represents the gold standard of cell therapy for cartilage repair with up to 16 years' follow-up and more than 80% of treated patients showing sustained improvement. As it has been suggested that the repair tissue of microfracture may not be durable, long term results would be needed to establish clinical superiority.

## *Stem cells as chondrocyte substitutes*

Articular chondrocytes are difficult to expand because of their limited proliferative capacity and their de-differentiation, resulting in the loss of their capacity to form cartilage when transplanted in vivo. Indeed, in some patients the repair tissue after ACI is poorly differentiated fibrocartilage and this could at least partly be due to the partial loss of the phenotypic stability of the expanded chondrocytes during in vitro culture.

Mesenchymal stem cells (MSCs) are easy to isolate and to expand in culture, they are chondrogenic and appear to be immune-privileged; hence they are attractive chondrocyte substitutes in an ACI procedure. These properties of MSCs would allow upscaling and generation of large batches of quality controlled cell preparations ready for allogeneic use, thus circumventing the limitations and patient-to-patient variability of autologous cell protocols. Preclinical and clinical studies are needed to compare MSCs with articular chondrocytes in an ACI procedure to see whether implantation of MSCs will result in a cartilage tissue that is as good and durable as the one following implantation of articular chondrocytes.

The use of 'universal stem cells' would be ideal. The availability of large batches of 'off-the-shelf' cell populations that are quality controlled for efficacy will enhance consistency of treatments while abating costs. Their availability will also eliminate the need for two operations and enable large scale production. This however poses obvious risks of rejection. There is evidence that stem cells such as MSCs from mismatched donors can be poorly immunogenic in vivo under specific conditions. However, the differentiation into a mature phenotype of the implanted stem cells is likely to result in the loss of the immunological privilege, with consequent rejection. Strategies to safely overcome the immunological barriers are intensely pursued.

## *Pharmacological targeting of endogenous joint stem cell niches*

The joint environment is a rich source of stem cells, which can be derived from synovial membrane and fluid, periosteum, bone marrow, and even the articular cartilage itself. Therapeutic approaches could target the endogenous resident stem cell pools to trigger and enhance joint surface regeneration. A key question, as yet unanswered, relates to the location and modulation of stem cell niches in the joint. A priority in the biomedical community is therefore the identification and characterisation of the stem cell niches in the joint and the investigation of how signals in the niches are coupled to functional events and related outcomes in joint homeostasis, remodelling and repair. Such knowledge will instruct the development of novel therapies to endorse joint tissue regeneration via endogenous stem cells. The increasing availability of small molecules and the development of spatio-temporally controlled delivery systems make modulation of microenvironments an attractive approach for joint surface regeneration.

## *Conclusions and perspectives*

Transplantation of bone marrow and haematopoietic stem cells in haematology best illustrates the success of a cell therapy that has evolved with the increasing understanding of cell phenotypes, functions, and niches. Classical ACI has evolved into second and third generation ACI with the use of 3D-matrices. It is hoped that in the near future, novel strategies for biological joint resurfacing will be developed in parallel to refinements of ACI- and microfracture-based treatments. Location, size

and depth of the lesion, status of the surrounding cartilage and the other joint tissues will provide guidance in underpinning the optimal clinical indication for each of the many approaches to joint surface defect repair, ranging from ACI (with chondrocytes, MSCs or other stem cell types) to pharmacological targeting, by using drugs, of the joint stem cell niches to influence their biological behaviour in order to restore joint homeostasis and effectively prevent OA.

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**References**

Brittberg, M., Lindahl, A., Nilsson, A., Ohlsson, C., Isaksson, O. and Peterson, L. (1994) Treatment of deep cartilage defects in the knee with autologous chondrocyte transplantation. *N.Engl.J.Med*. **331**, 889-895

## A Day in the Life of a Scientist in a Regional Burns Centre

I am a clinical scientist working in a small department in a West Midlands Hospital (University Hospitals Birmingham NHS Foundation Trust) that was established over 25 years ago to provide the regional Burns Centre (adult and paediatric) with a skin cell culture service. The department used to consist of a small laboratory and was unattractively, albeit accurately, referred to as "The Skin Lab". The department, now less graphically named UHB's Tissue Services, ensures all Trust activities using human tissue to treat patients are compliant with the requirements of the Human Tissue Authority, as well as providing a supply of cultured skin cells to Burn Surgeons in the UK. I am the registered Designated Individual for the Trust's Human Tissue Authority Licence for Human Application.

Our work involves the procurement, storage, processing and use of autologous and allogeneic skin, skin cells and skin substitutes in the treatment of burns, the storage of allogeneic bone and tendon used in orthopaedic and trauma surgery, the procurement of autologous and allogeneic peripheral blood stem cells and the storage of allogeneic liver vessels; any Trust activity that requires HTA (Human Application) licensing is managed by my team.

The department has a Clean room facility (which we still fondly refer to as The Lab, despite it bearing no resemblance to any traditional laboratory) where cultured skin cells are produced and also a Tissue Bank Facility for the storage of cryopreserved and refrigerated tissues.

Like everyone working in the NHS there really is no typical day; this makes for an interesting working life.

The Regional Burns Centre is served by a team of seven Consultant Plastic and Burns surgeons. From June 2010 the Service has been provided from a 15-bedded Centre in the new Queen Elizabeth Hospital Birmingham. This includes seven heated shock rooms and four isolation rooms. The Burns theatre and 100 bedded critical care unit (with two purpose-built shock rooms with complete body showering facilities) are adjacent to the Burns Centre.

The surgeons use cryopreserved skin in the management of burns injuries and over 75,000 $cm^2$ of cryopreserved skin is used each year by this Trust. Once a severely burned patient has been admitted to our hospital, the surgeons carry out early excision of the burn, generating an immediate need for substantial amounts of cryopreserved skin. We have our own licensed storage facility to maintain stocks of cryopreserved skin for immediate use; all cryopreserved allograft skin is provided by NHSBT's Tissue Services in Liverpool.

Most large burns (>30% total body surface area, Total Body Surface Area (TBSA)) admitted to our hospital are treated routinely with autologous cell culture therapy: a small sample of the patient's skin is sent to the laboratory, skin cells, namely keratinocytes, are extracted and cell numbers are rapidly increased to provide surgeons with high density suspensions of activated keratinocytes, which are used to *re-epithelialise* wounds.

Cell culture is a bit like gardening and a bit like cooking! I have often described the ability to culture

human cells consistently well as a capricious art, rather than an exact science; experience leads me to believe there are those that can and those that can't and although the recipe for success may *seem* easy enough to follow, the end results are by no means guaranteed. I have a team of highly skilled staff working in the cell culture laboratory that not only provide a consistently high quality cell culture service but we also generate a steady stream of allotment produce, fabulous cakes and sumptuous savouries in our spare time. Cell culture, gardening and cooking abilities seem to go hand in hand!

Today, my first job is to ensure that all systems are go: are all facilities working and all tissues and cells stored correctly? The next check is on overnight activity; have there been any admissions overnight that will require tissues or cells? Have any of the stock tissues been used needing replacing and has all use been appropriately recorded?

We have used a substantial amount of cryopreserved skin in the early hours and so replacement stocks are ordered from NHSBT in Liverpool. Some of the cryopreserved skin was rejected by the Consultant. It transpires that some of the cryopreserved skin is tattooed and the surgeon rejected this skin on the grounds that cryopreserved skin has been known to "take" and so use of tattooed skin was not considered appropriate. The team have never been presented with this situation before and were not aware that it might occur. There is no quality or safety issue however, and after debate, the surgeons' consensus is not to use tattooed skin in our Centre and so I am asked by the Clinical Service Lead to discuss this issue with staff from NHSBT's Tissue Services. In the context of a large increase in the incidence of skin tattoos in the population there had been a policy for tattooed donors of skin to be accepted depending on skin stocks and as long as the tattoos were not identifiable and were not extensive. This has resulted in another review of the situation within NHSBT Tissue services which will be discussed, as it has in the past, with the burns surgeons around the country through the NHSBT Burns Skin Forum. I relay the information back to the surgeons in a report for them to digest and comment on. This is something that cannot be resolved today but by having stocks of skin in our own Trust facility, the surgeon was able to carry on with his procedure and the rejection of some packs of skin has had no negative clinical impact.

I review on-going clinical work with the laboratory team and ensure that cultures are going to be ready for requested applications dates. We have four different patients' cells being cultured at present and the laboratory team are busy.

Just before lunch, I receive a telephone call from the paediatric burns team regarding a new patient. We are asked to culture cells for the patient and also to send some cryopreserved skin over to the operating theatre; skin and blood samples are on their way over to the laboratory. The management of this new patient's cells is discussed with the team.

UHB NHS Foundation Trust is a large organisation which has recently undergone the first of several phased moves into the city's first new acute hospital in seventy years. The Burns Centre moved in its entirety in June, however the new clean room facility is not ready; UHB Tissue Services are still located in our original facility for the time being. Some of my time is spent planning and co-ordinating aspects of the different moves we are linked with, as well as overseeing the build and commissioning of the new clean room. I spend a couple of hours inspecting the new clean room with the Project Director and his entourage. The facility is so specialised it is essential that it meets the specification. I then take information from this meeting back to my old, familiar and comfortable office and continue planning the commissioning process and subsequent move. Moving into the new facility requires new licence applications (and several trees worth of accompanying paperwork) and so I settle down at the computer and continue work on the new Quality Manual for the new facility; another day passes and I haven't been anywhere near a pipette or a Petri dish! Maybe I can get into the laboratory tomorrow!

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# Cardiac Repair in Patients with Coronary Heart Disease: The Use of Bone Marrow Derived Cell Preparations

## *Why do we need cardiac stem cell therapy?*

Cardiovascular disease is the leading cause of death in both developed and developing countries. The heart shows an inadequate ability to repair itself after myocardial infarction and other forms of heart disease, resulting in impaired contractile function and cardiac enlargement, the result of which is the development of heart failure. Heart failure is a growing problem in the UK, affecting ~900,000 individuals at a cost to the NHS of ~£680 million per year. Heart failure carries a one year mortality of ~40% which is worse than most cancers. Improvement of cardiac function through delivery of novel cellular therapies with the aim of replacing lost cells has therefore been a major strategic focus for the emerging field of regenerative medicine.

Functional benefits of cell therapy for cardiovascular applications may arise from induction of growth of new blood vessels (angiogenesis), regeneration of heart muscle cells (cardiomyogenesis), improved survival of heart tissue (cardioprotection) or mechanical interstitial support. The former three modes of benefit may result from site-specific trans-differentiation of administered cells or by secretion of paracrine factors that stimulate endogenous repair or protective mechanisms. On this basis, improvements in regional myocardial perfusion, contractile function and adverse ventricular remodelling would be predicted. The additional mechanical interstitial support provided by cell administration may itself modulate adverse ventricular remodelling. These potential benefits need to be weighed against the potential toxicity from cell therapy, such as accelerated atherosclerosis, increased arrhythmia risk, and local or ectopic tumour formation or calcification.

## *Unselected Bone Marrow Mononuclear Cells*

Phase I trials of unselected bone marrow mononuclear cells (BMMNC) in patients with acute myocardial infarction and ischaemic cardiomyopathy, delivered by intracoronary infusion and endomyocardial injection, respectively demonstrated procedural safety for the delivery methods and no short term toxicity. The uniformly observed benefits in these non-randomised, open label, non-placebo controlled studies appeared independent of cell dose and timing of administration. The magnitude of benefit could not be replicated in first generation randomised clinical trials in patients with acute ST elevation myocardial infarction (STEMI). Recent meta-analyses suggest that BMMNC administration in patients with acute STEMI is associated with an ~3% increase in Left Ventricular Ejection Fraction (LVEF), ~5mL reduction in left ventricular end systolic volume and ~3.5% reduction in myocardial infarct size compared to placebo. These benefits are modest and have not translated into an overall reduction in major adverse clinical events. However, local BMMNC administration offers a trend to greater benefit in those patients with greatest impairment of ventricular function.

BMMNC have also been administered to patients with chronic myocardial infarction and ischaemic cardiomyopathy, either by intracoronary infusion or direct myocardial injection. The latter can be performed at the time of Coronary Artery Bypass Grafting (CABG) or using a percutaneous endomyocardial injection catheter. Open label studies in this patient group have demonstrated significant improvements in LVEF, myocardial perfusion and cardiopulmonary exercise testing. These studies enrolled small numbers of patients. In surgical studies, it has been difficult to distinguish the effects of injected cells from the confounding effects of revascularisation on changes in regional and global ventricular function. The largest randomised studies to date have shown that intracoronary infusion of BMMNC into the artery supplying the most dysfunctional myocardial segment produced ~3% increase in LVEF at three month follow-up. Interestingly either intracoronary or myocardial injection into chronically infarcted regions at the time of CABG produced no increase in either regional or segmental ventricular function. A number of ongoing investigations are evaluating the potential benefits of BMMNC in ischaemic cardiomyopathy.

## *Selected Bone Marrow Mononuclear Cells*

Investigators have reported the use of $CD133^+$ and $CD34^+$ cells, for the treatment of patients with both acute STEMI and refractory angina. This is based upon pre-clinical literature which suggests that these cells can:

- Differentiate into an endothelial cell and cardiac myocyte phenotype;
- Promote neovascularisation through paracrine mechanisms;
- Improve left ventricular function in animal models of myocardial infarction.

In an open label study, $CD34^+$ $CXCR4^+$ progenitor cells selected from bone marrow were administered by intracoronary infusion to 80 patients with acute STEMI involving the left anterior descending artery. There was no significant improvement in the $CD34^+$ $CXCR4^+$ cell treated group compared with those receiving either unselected BMMNC or no cell therapy. Furthermore, cell therapy was not associated with attenuated adverse left

ventricular remodelling. In addition, selected $CD34^+$ cells have been administered by percutaneous endomyocardial injection in 24 patients with refractory angina. This study reported no significant safety concerns, and improved angina frequency and exercise capacity in the group receiving $CD34^+$ cells. The final published results of the randomised Phase II study are awaited.

$CD133^+$ progenitor cells ($12.6 \pm 2.2x10^6$) from the bone marrow, have been infused into infarct-related arteries in 19/35 patients, ~12 days after acute STEMI. Patients receiving $CD133^+$ cells had improved segmental wall motion and regional myocardial perfusion, but had an increased risk of stent occlusion, in-stent restenosis or development of de novo coronary lesions. Other feasibility studies of intramyocardial injection of $CD133^+$ cells at the time of CABG have been reported, with larger trials currently underway.

## Mesenchymal Stem Cells

Mesenchymal stem cells (MSC) are a self renewing population of cells in bone marrow which can transdifferentiate into several cell types, including cardiac myocytes, and can be stimulated home to infarcted myocardium via the CXCL12/CXCR4 chemokine axis. MSC are potent sources of angiogenic and immunomodulatory cytokines. Furthermore, MSC may be used for allogeneic administration without concomitant immunosuppressive therapy, due to the absence of major histocompatibility and costimulatory antigens on the cell surface. A Phase I double blind placebo-controlled dose escalation safety study of $0.5 – 5x10^6$ MSC administered intravenously to patients with acute STEMI, showed no deterioration in pulmonary function tests or increased ventricular ectopy. In a subset of patients (20 MSC treated v 14 placebo treated), a significant and sustained increase in LVEF in the MSC treated group ($5.2 \pm 8.5\%$ v $1.8 \pm 6.7\%$) was reported, as well as attenuated adverse left ventricular remodelling. However, these data should not be over-interpreted, as placebo-treated subjects in the CMR subgroup appeared to have larger infarctions and in the analysis of the entire study group, echocardiographic measurement of LVEF and exercise time were no different in the MSC and placebo treated groups. Overall these data are promising and further investigation of allogeneic administration of MSC is warranted.

Multipotent adult progenitor cells (MAPC) are another stromal population, which are capable of self renewal and differentiation into cells of all three germinal layers. Initial experiments in a swine model showed that intramyocardial injection of $50 \times 10^6$ allogeneic MAPC into the infarct border one hour after the application of coronary ligation resulted in improved regional and global contractile function by CMR. Interestingly, only ~3% and ~2% of the cells developed an endothelial or cardiomyocyte phenotype suggesting that the majority of functional benefit may be achieved through a paracrine mechanism. MAPC may be considered for clinical evaluation pending future successful efficacy and safety studies, particularly with respect to cytogenetic abnormalities that may be associated with prolonged expansion under stringent culture conditions.

## Progenitor Cell Mobilisation

Cytokine mobilisation of progenitor cells may confer haemodynamic benefits in animal models. A recent meta-analysis concluded that G-CSF administration conferred no functional benefit to patients with acute STEMI. Recent studies suggest that combination therapy with AMD3100 (plerixafor, antagonises the CXCR4 chemokine receptor) and Vascular Endothelial Growth Factor (VEGF) may selectively mobilise endothelial and stromal progenitor cells while suppressing the release of haematopoietic progenitor cells and neutrophils from bone marrow, and warrants further evaluation.

## Future Challenges

On the basis of the available data, bone marrow derived cell therapy should not be considered part of routine clinical treatment for patients with cardiovascular disease. Many unresolved issues remain. The optimal cell preparation for each clinical application remains undetermined. It cannot be assumed that a single cell preparation will be equally efficacious for all clinical applications, and different cell preparations may have varying toxicity profiles. Indeed, it is unclear if administration of a highly selected cell population is preferable to a heterogeneous unselected or combination cell product. Other unresolved issues include the optimal number of cells to be delivered, timing of cell administration, route of administration, importance of growth factor preconditioning of cellular products prior to administration, effects of *ex vivo* cell expansion and prolonged cell culture prior to administration, and the use of allogeneic rather than autologous cell preparations. Numerous small scale clinical trials are being conducted worldwide to address some of these issues, but few are likely to be sufficiently powered to provide definitive conclusions.

Based on current experience, how might we improve the evaluation of the next generation of cellular therapies, such as resident cardiac stem cells and induced pluripotent stem cells, which can be differentiated into heart muscle with greater efficiency than bone marrow derived cell preparations? In the future, more robust pre-clinical experimentation using clinically relevant cell preparations, disease models and endpoint assessment may improve identification of cell preparations which

translate to significant clinical benefit in patients. Furthermore, randomisation and blinding should be implemented at the earliest stages of clinical evaluation. For autologous cell products, biological assays of an aliquot of the administered cells are highly desirable in order to understand the contribution of variation in biological potency of a cell product, which can clearly vary from individual to individual, to any observed variability in trial efficacy and safety endpoints. This may require the development of "potency" based release assays for cellular products and clearly poses significant challenges to clinical good manufacturing practice protocols for processing of cellular products for cardiovascular applications.

## *Conclusions*

Many obstacles remain along the path to successful routine clinical application of cardiovascular cell therapy. Much has been learned from the initial clinical experience, but perhaps the emphasis should now focus on further pre-clinical work to evaluate the efficacy and toxicity profiles of next generation cell preparations, rather than moving forward to additional clinical trials. This will be a lengthy and complex process, the success of which will require an integrated multi-disciplinary collaboration between basic scientists, cardiologists, cardiac surgeons, haematologists, cell-processing experts and industry.

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### References

Wollert KC, Drexler H (2005) Clinical applications of stem cells for the heart. *Circ Res*, **96(2)**,151-163.

Kang S, Yang YJ, Li CJ, Gao RL (2008). Effects of intracoronary autologous bone marrow cells on left ventricular function in acute myocardial infarction: a systematic review and meta-analysis for randomized controlled trials. *Coron Artery Dis*, **19(5)**, 327-335.

Martin-Rendon E, Brunskill SJ, Hyde CJ, Stanworth SJ, Mathur A, Watt SM (2008) Autologous bone marrow stem cells to treat acute myocardial infarction: a systematic review. *Eur Heart J*, **29(15)**, 1807-1818.

Tendera M, Wojakowski W, Ruzyllo W, *et al* (2009) Intracoronary infusion of bone marrow-derived selected $CD34^+CXCR4^+$ cells and non-selected mononuclear cells in patients with acute STEMI and reduced left ventricular ejection fraction: results of randomized, multicentre Myocardial Regeneration by Intracoronary Infusion of Selected Population of Stem Cells in Acute Myocardial Infarction (REGENT) Trial. *Eur Heart J*, **30(11)**, 1313-1321.

Assmus B, Honold J, Schachinger V, *et al* (2006) Transcoronary transplantation of progenitor cells after myocardial infarction. *N Engl J Med,* **355(12)**, 1222-1232.

Ang KL, Chin D, Leyva F, Foley P, Kubal C, Chalil S, *et al* (2008) Randomized, controlled trial of intramuscular or intracoronary injection of autologous bone marrow cells into scarred myocardium during CABG versus CABG alone. *Nat Clin Pract Cardiovasc Med,* **5(10)**, 663-670.

Losordo DW, Schatz RA, White CJ, *et al* (2007) Intramyocardial transplantation of autologous $CD34^+$ stem cells for intractable angina: a phase I/IIa double-blind, randomized controlled trial. *Circulation*, **115(25)**, 3165-3172.

Bartunek J, Vanderheyden M, Vandekerckhove B, *et al* (2005) Intracoronary injection of CD133-positive enriched bone marrow progenitor cells promotes cardiac recovery after recent myocardial infarction: feasibility and safety. *Circulation*, **112(9 Suppl)**, I178-183.

Stamm C, Kleine HD, Choi YH, *et al* (2007) Intramyocardial delivery of $CD133^+$ bone marrow cells and coronary artery bypass grafting for chronic ischemic heart disease: safety and efficacy studies. *J Thorac Cardiovasc Surg*, **133(3)**, 717-725.

Hare JM, Traverse JH, Henry TD, *et al* (2009) A randomized, double-blind, placebo-controlled, dose-escalation study of intravenous adult human mesenchymal stem cells (prochymal) after acute myocardial infarction. *J Am Coll Cardiol*, **54(24)**, 2277-2286.

Zeng L, Hu Q, Wang X, *et al* (2007) Bioenergetic and functional consequences of bone marrow-derived multipotent progenitor cell transplantation in hearts with postinfarction left ventricular remodeling. *Circulation*, **115(14)**, 1866-1875.

Zohlnhofer D, Dibra A, Koppara T, *et al* (2008) Stem cell mobilization by granulocyte colony-stimulating factor for myocardial recovery after acute myocardial infarction: a meta-analysis. *J Am Coll Cardiol*, **51(15)**, 1429-1437.

# Knowledge is Power: The ABC Experience

For centuries the adage "knowledge is power" was meant as a prescription for dictators to enslave their people in ignorance. Even today some governments try to restrict citizen access to certain books and websites. Unfortunately, it is also not unusual for the elite "in the know" in an organization to limit the flow of information to its constituents so they seem that much smarter. How 1970s is that?

America's Blood Centers (ABC) is an alliance of non-profit blood programs that provide half of the US and one-quarter of Canada's volunteer donor blood supply. That's about 10 million red blood cells, plus platelets and plasma to support nearly three million patients. Many ABC members are also involved in marrow donor recruitment, cell therapies and tissue services. Increasingly, hospitals are outsourcing their cross-matching and transfusion services to ABC centers.

Since 1962 ABC has believed the best place for information was in the hands of the operators, the users. This may seem quaint today, but in the 1960s when ABC started a weekly newsletter, it was downright revolutionary. In those days, the staff of ABC gathered clippings from newspapers, memos, government publications, anything having to do with blood collection and processing. The cut-and-pasted newsletter (copied on the newly available Xerox machines) soon attracted readers outside the ABC membership. By the early 1980s, blood centers and hospitals around the US and in many countries waited expectantly for the latest and greatest from the weekly *CCBC Newsletter*. (In 1996 the Council of Community Blood Centers or CCBC changed its name to America's Blood Centers).

In the 1980s, AIDS spread through the blood supply around the time that mainstream news media went 24/7. Unfortunately, bad decisions were made by blood providers in several countries, either for a lack of good information and in a few cases despite the latest knowledge. Scandals in several countries resulted in totally reorganized blood programs, but no country was exempt from a new way of looking at transfusion risks going forward. During that period, the *CCBC Newsletter* became the chronicle of AIDS in the blood supply detailing discussions from early 1980's meetings and actions taken in the US and elsewhere (where known) to help staunch transmissions.

Today no country is an island. Regulators and blood programs are globally in touch minute-by-minute on any new development in blood safety, although transparency in providing such information to the end-user is still not ideal. No country can make a blood safety recommendation without every other country evaluating its applicability.

Starting in the 1980s, ABC/CCBC kept its weekly chronicle but also began sending daily bulletins – there was just so much information coming out; first by postal mail, then by fax in 1990s (so called "fax attacks" because every morning ABC members had upwards to an inch of papers printed in their fax machines). Today we send out literally thousands of targeted messages annually (by email and between blood centers by ListServ – sort of a managed single topic group email or blog) to keep our members informed and so they can implement the latest and greatest, and also work with their hospitals to make informed decisions on risks and benefits of new safety threats and interventions.

Sometimes it is hard to keep up and sort out what's what. The *ABC Newsletter* has now become a weekly digest for tens of thousands of blood workers and hospital personnel around North America and again in many countries. See the latest edition at: **http://www.americasblood.org/download/File/newsletter_sample.pdf.**

ABC also annually puts together at least half a dozen monographs to the hospitals served by its members. These single-topic, two-pagers are written by medical and healthcare experts on transfusion and blood-related business topics, and peer reviewed and referenced with the latest information prior to release to hospitals through ABC members. "Blood Counts" deals with the business aspects of blood banking while "Blood Bulletin" gives physicians, nurses and technologists the latest on blood usage topics. Both are available on the public side of the ABC website (**www.americasblood.org** under publications).

What's ahead? Well, in Europe, and especially in the UK, we know controlling inappropriate blood use has been a hot topic for well over a decade, but the subject has just caught fire in the colonies. ABC has adopted the excellent blood inventory management tool used by most hospitals in the UK (VANESA or the Blood Stocks Management Scheme) and linked it to hospital lab information systems for automated data transfer and tracking. We also have automated a blood utilization clinical monitoring and benchmarking program developed in Finland (called VOK, standing for Optimal Blood Use in Finnish).

Within months these programs (collectively called AIM in the US) will be installed in hundreds and, by the end of 2011, in thousands of US hospitals generating reams of data on best practices, adverse and optimal patient outcomes, trends and new ways of treating patients with transfusions. Daily ListServs between hospital users may be the best way to communicate this information on a

timely basis in a way that empowers everyone. However, best practices and the most useful information will likely be targeted for analysis in a weekly or monthly digest to hospital users.

AIM will likely be installed in other countries (e.g. Flanders and the Netherlands have already expressed interest), so best practices and new information may start emerging from countries around the world for use by hospital transfusion committees, and others responsible for appropriate blood use.

What's been true for centuries is still true today: knowledge is power; the power to make informed choices. Today's problem is managing too much information and truly gleaning "knowledge" from often too much background noise. Just giving people information can be helpful, but showing them how it can be used as a tool to improve patient care remains critical and also labour intensive. For ABC, information and knowledge sharing and management with our blood center members has been our strength and our reputation. The ultimate beneficiaries have been the hospitals and the transfusion recipients all of us serve.

**Jim MacPherson**
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**America's Blood Centers**
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## The World Health Assembly Resolution on Organ and Tissue Transplantation

Organ and Tissue Transplantation is not really central to the business of the World Health Organisation or the World Health Assembly (WHA). Amongst the toll on human life it is the communicable diseases that loom largest. The work of the WHA thus primarily involves joint efforts, directed for example at eradication of smallpox and polio, or to tackle HIV with the United Nations parent body. Blood has of course been addressed extensively through this agenda because of the risks of disease transmission and the need to provide blood for transfusion as a basic healthcare service. Why then has there been a veritable proliferation of resolutions from the assembly on transplantation? WHA 40.13 (1987) and WHA 42.5 (1989) noted concern over commercial trade in organs and the need for global standards for transplantation. The first WHO Guiding Principles on Human Organ Transplantation were endorsed in resolution WHA 44.25 in 1991. In 2004, 13 years later WHA 57.18 asked the WHO Director-General "to continue examining and collecting global data on the practices, safety, quality, efficacy and epidemiology of allogeneic transplantation and on ethical issues, including living donation, in order to update the Guiding Principles on Human Organ Transplantation". Here there is the clue to its importance – ethics and safety, or to be more precise: concern over unethical and unsafe practices.

The 2010 World Health Assembly resolved in WHA 63.22, to endorse the revised Guiding Principles: the work of the WHO that had taken from 2005 to 2009 to complete; that had been considered by the WHO Executive Board in 2009; and that had been dropped from the agenda of WHA 62 in 2009 when the world caught 'Swine Flu'. The wheels of international diplomacy and debate revolve at an almost imperceptible rate. For those who had been through every step of the 7 or 8 years from initiation to resolution, it was a painful experience to watch the WHA in May this year as they moved at glacial speed down an agenda filled with complexity and discord. One of the two committees became bogged down in determining the mechanism for appointment of the Director General and there were times when there was uncertainty as to whether our agenda item would ever be reached. At the WHA ''It is never over until it is over", since any country delegate can intervene and request amendments which can, by the stroke of a pen, turn the meaning of a resolution on its head. On this occasion, late on Friday at the end of the assembly, many countries stood to register their support for the resolution. Australia changed one word in the resolution – from 'welcomes' to 'endorses' and in that word, and in the unanimous subsequent agreement of the WHA, every country in the world represented at the World Health Assembly bound themselves to the revised Guiding Principles.

The intent of the revised Guiding Principles is to reinforce the previous advice on donation and transplantation as well as provide new guidance to governments with respect to transparency, to allocation, to regulation of safety and regulatory oversight of transplantation and to detailed issues such as encouragement for globally consistent coding systems for human cells, tissues and organs to assist in traceability. The Resolution also asks the WHO Director General to undertake actions which provide an ongoing workload for the WHO Secretariat. This can be interpreted as the governments of the world expressing concern about

organ trafficking, adverse events after transplantation, seeking analysis of global data on safety, quality, efficacy, epidemiology and lastly looking for international vigilance with respect to the ethics of transplantation practices.

The revised Guiding Principles include nine principles that have not changed significantly from the 1991 version and two new principles, numbers 10 and 11 (Boxes). Principle 10 focuses on safety, quality and efficacy, while 11 concentrates upon the twin concepts of transparency of practice and protection of the privacy of individual donors and recipients. These principles thus place responsibility on governments to adjust their regulatory systems over the years ahead.

**Guiding Principle 10:**

High-quality, safe and efficacious procedures are essential for donors and recipients alike. The long-term outcomes of cell, tissue and organ donation and transplantation should be assessed for the living donor as well as the recipient in order to document benefit and harm.

The level of safety, efficacy and quality of human cells, tissues and organs for transplantation, as health products of an exceptional nature, must be maintained and optimized on an ongoing basis. This requires implementation of quality systems including traceability and vigilance, with adverse events and reactions reported, both nationally and for exported human products.

**Guiding Principle 11.**

The organization and execution of donation and transplantation activities, as well as their clinical results, must be transparent and open to scrutiny, while ensuring that the personal anonymity and privacy of donors and recipients are always protected.

The work of the WHO has thus just started anew. There is the dissemination of the Guiding Principles – commencing on the website **http://www.who.int** and published in the journal *Transplantation* in August. The work of implementation of coding systems will be arduous and more complex than it sounds. Interplay between the regional supra-national organisations will be interesting – but has got off to a good start with a joint EU/WHO/Transplantation Society meeting in Madrid. Data collection and analysis is a work in progress to be found on the WHO global observatory (**http://www.transplant-observatory.org**). The challenges to the goal of ethical practice are however to be found in far too many countries – China, Peru, The Philippines, India, Egypt and Sri Lanka amongst others. But can we be sure that all is well at home – perhaps the first place to attend to is one's own back yard? It will pay us all to examine the WHO Guiding Principles carefully and thoughtfully and then to consider how well our own practices fit the global norms represented in this document.

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# Historical Perspective of Blood Transfusion in War-Time Britain

By 1939 much remained to be learnt about transfusing blood safely. The *total* British military dead during World War 2 was over 350,000; and at least 60,000 civilians and 1,200 'Home Guard' were killed. Given that non-fatalities outnumbered fatalities, the demand on transfusion services was immense, so the learning curve was steep. Nevertheless, many tens of thousands were saved by transfusion.

Although Landsteiner had indicated that his discovery of the ABO system could improve transfusion safety, knowledge was slow to spread. His work was discovered independently by Janský and by Moss; each gave a different Roman Numerical nomenclature which plagued transfusion practice (see table). In 1940 Goodman, Britain's representative of the League of Nations' Health Committee (LoN), warned of the continued dangers of labelling blood for transfusion by the Moss system.

**Equivalent nomenclatures for ABO Blood Groups**

| Landsteiner 1902 | Janský 1907 | Moss 1910 | LoN 1928, 1940 (*von Dungern, 1911*) |
|---|---|---|---|
| A | II | II | A |
| B | III | III | B |
| C | I | IV | O |
| 'no particular type' | IV | I | AB |

Citrate-based anticoagulants, introduced during WW1 sometimes with added sugar, allowed collection and transfusion to be separated in space and time. This encouraged 'volunteer panels', first set up by Percy Oliver of London in the early 1920's. However in 1939 'the ideal anticoagulant' was 'still to be found', optimum chemical formulations and blood:anticoagulant ratios remained uncertain.

At first most donations came from 'universal donors' (group 'O IV'), but in time non-O blood became used. Ipswich, led by Dr E Biddle, started a systematic service in 1928 and by 1938 had collected 150 donations within 10 months, 66% of which were group O, 58% coming from volunteers (42% being recipients' relatives). Pint-bottles with 50ml of citrate-only solution received up to 500ml donor blood: about 10% of donors had a 'mild reaction', and donation could be repeated after a month. The average collection-to-transfusion time was 4.4 days (maximum 14); blood was poured through a gauze filter into another bottle and warmed toward body temperature before administration (this 'warming' recommendation was universal). About 5% of patients had a mild rigor but no worse.

The Army Blood Transfusion Service (ABTS) was established at Southmead Hospital, Bristol, in 1938. Sir Lionel Whitby, a WW1 veteran, was put in charge assisted by Lt Geoffrey Tovey. In its first year the ABTS processed over 33,000 donations, six times more than the busiest civilian service, but only white Anglo-Saxon soldiers of group O were selected. Concentrating on France until after Dunkirk, in 1943 it supplied over 100,000 units of plasma to the 8th Army in North Africa.

Also in 1938, the Medical Research Council (MRC) established metropolitan 'depots' at Luton, Maidstone, Worcester Park (Sutton), and Slough: with a busy nursing and admin staff, these collected and stored blood for delivery to 'sector hospitals' throughout SE England. Each depot was to have 1,000 bottles and 20,000 volunteers on a donor panel; they were led respectively by Drs Brewer, Maizels, JO Oliver (succeeded by JF Loutit in 1941) and Janet Vaughan. Vaughan, of Hammersmith Hospital, a cousin of Virginia Woolf and author of a textbook on anaemia (1936), had volunteered with Dr Duran Jorda's BTS at Barcelona, established during Spain's tragic civil war, 1936-9.

Elsewhere, civilian services had to do their best. Protests over 'London bias' (and petrol rationing) came from Merseyside, Leeds, Wolverhampton, Portsmouth and Ipswich. Scotland and Wales got their own services. Attempts to temper enthusiasm warned of transmitted diseases; one syphilologist recommended thorough physical examination of all donors (mostly young men). In Edinburgh a plasma separation plant was built, in a basement at the Royal Infirmary, by Andrew Crosby (who later became 'Chief Technician').

The 'phoney war', the quiet period until the Fall of France in May 1940, gave time for the depots to prepare for the *Blitz* (September 1940 to May 1941). After November the Provinces (South Coast, Bristol, South Wales, Merseyside, Tyneside, Glasgow and an ill-prepared Belfast) were targeted. Non-military cities such as Bath were bombed in mid-1942 as reprisals for Allied attacks on Germany; another wave started in December 1943 and V-bombs fell on London in late 1944.

Persisting poor preservation hampered distant delivery at home and overseas. One approach was to ship the more stable plasma separated from unused blood (at Sutton, PL Mollison successfully transfused the remaining red cells re-suspended in glucose/saline). In an extra-ordinary humanitarian gesture, Dr Charles Drew of New York supplied thousands of dried plasma units to Britain in 1940. But the Canadians also shipped several hundred units and the British soon developed their own techniques, so much American plasma was returned.

Maybe related was the increasing recognition of 'serum hepatitis' transmission – at first from vaccines containing traces of human serum but also by blood transfusion, especially plasma. Nevertheless, plasma transfusion for traumatic shock (and burns), especially prior to whole blood, was widely praised as a "life-saver", the occasional late-onset jaundice being largely ignored.

In 1942 Mollison devised an improved glucose-rich anticoagulant-preservative ("ACD-A"). An inferior formulation of 180ml with double its volume of blood had often been used, and blood was often observed to be brown after two weeks (*Dr John Perrin*, 1973). ACD-A's lower volume (120ml) allowed up to 430ml to be collected into the modified pint-sized milk-bottle container (plus 10ml for the sample-testing tubes) and stored cold for three weeks. A steel-wire loop from a metal band clipped at the base of the bottle allowed it to be inverted so that blood could be delivered to the recipient through a rubber tube. It took many variations before this simple "MRC bottle" design took hold. Collecting more than 440ml increased the faint rate and a minimum donor weight of eight stone (50 Kg) was set although 400ml was still collected from donors weighing between seven and eight stone.

The 'Rhesus factor', discovered in 1939, led to important serological developments. By 1944 Race had observed apparently paradoxical production of anti-Rh by Rh **positive** individuals – the 'CDE' concept of multiple but linked Rh genes only emerged (from Fisher) in 1945. However, knowledge of the Rh factor was already enabling babies with 'erythroblastosis foetalis' (allo-immune haemolytic disease) to receive appropriate Rh negative blood, saving several lives although more were to be saved by the post-war development of exchange transfusion (which combined the infusion of good quality non-reactive red cells with removal of excess bile pigment and the haemolysis-prone cells from the baby) and use of plastic delivery sets (which were easier to use and clean, and had a much lower incidence of transfusion-associated non-specific febrile reactions). It was also realised that actively bleeding babies with the very different 'haemorrhagic disease' could benefit from vitamin K injections, but blood transfusions were better and quicker to take effect.

In 1945, while Race was unravelling the Rh system, Robin Coombs developing the ground-breaking anti-globulin test and Walter Morgan steadily improving ABO chemistry understanding, about 200,000 units of donations were collected in Britain. With peace, demand halved; but post-war developments, which included transferring the wartime organisation to the new NHS, soon caused activity to rise way beyond – leading to the post-war 'golden age'. These included clarifying the ramifications of the Rh system and the vast expansion in the science of the multiple blood group systems generally (including forensic applications), improved compatibility-testing techniques in blood bank laboratories, the introduction of component therapy (including for haemophiliacs), improving standards generally (including donor care) and support for increasingly complex medical procedures such as heart surgery (including for babies), transfusion support in leukaemias and – later – organ and stem cell transplantation. Nevertheless, we owe a huge debt to the wartime pioneers of transfusion and their work, often conducted under unimaginably great difficulties.

*Many details are to be found in contemporary issues of the BMJ and Lancet.*

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## Obituary: Professor Kazimierz Ostrowski (1921-2010)

Professor Kazimierz, Ludwik Ostrowski one of the most creative scientists in Poland, passed away on January 9th 2010.

Professor K. Ostrowski was born on October 24th 1921 in Lwow. During World War II, thanks to friends he moved to Warsaw. After the war he enlisted as a student in the Medical faculty of the University of Warsaw and in 1949 became a physician. In 1948 he was engaged by Professor Juliusz Zweibaum to work in the Department of Histology and Embryology as a researcher and received his doctorate in 1951. In 1955 the position of "docent in histology" was granted to him. In 1956 he became the head of the Department of Histology and Embryology receiving the title of Professor of Medical Sciences in 1965. He was a head of the Department until 1992, when he retired.

The Professor's lectures were attractive to students and to physicians alike. He understood the needs for modern education of doctors and he taught some elements of

cell biology. In the sixties he introduced molecular biology and physiology, "cytophysiology" into the medical education and all medical schools in Poland followed suite. Professor edited the first handbook of cytophysiology.

An important part of his activity concerned objective assessment of histological methods. He established international contacts with Pearse from London, Hancox from Liverpool, Sandritter from Frankfurt, Barnard from Buffalo, Krompecher from Debrecen, Aron from Strasbourg, Czyba from Lyon, Wegman from Paris, Tchakaroff from Sofia and many others. The Professor was involved in investigation of heterotopic osteogenesis and in his last years was interested in expression of the genes of some morphogenic proteins (BMPs) in bone tissue and found that human expression of BMP genes were different in various parts of the skeleton.

In 1963 Professor Ostrowski collaborated with orthopaedic surgeons regarding preservation and storage of bone allografts for reconstructive surgery. Under his supervision the preparation of grafts for hospitals and clinical wards began in Warsaw. New multi-tissue tissue bank laboratories were created in Katowice (1968) and Kielce (1975), heart valves banks in Warsaw and Kraków (1980) and in Zabrze (1992). Ophthalmologists in collaboration with Professor Ostrowski, set up cornea banks in Lublin (1992) and in Warsaw (1995). He chaired the Commission of Transplantology of the Polish Academy of Sciences that organized over twenty conferences on tissue and organ banking and transplantation. His activities resulted in new directions for research. Radiation sterilization of grafts was implemented but initially it was not clear whether this might evoke appearance of free radicals in transplanted tissues. Investigation of irradiation of tissue components and of immunological reactions to transplanted bone were undertaken.

In 1970 Professor Ostrowski became director of the newly created Institute of Biostructure, in the Department of Transplantology & Central Tissue Bank. In 1975 the ministerial program (MZ-XIV) for cell and tissue preservation and storage was established under his direction. In 1987 he became Chairman of the Team of Specialists in Organ and Tissue Procurement and Transplantation, established by the minister of Health and Welfare (in 1995 reorganised as the National Transplantation Council). This resulted in numerous training conferences on transplantation and tissue banking, the distribution of 200,000 allografts, over 40 publications on tissue banking (including the first monograph in Polish about tissue banking in 1964), three international meetings (International Symposium in Jabłonna 1977, 2nd World Congress of Tissue Banking in Warsaw 1999, European Congress on Tissue Banking in Kraków 2009), and the development of regulation of organ and tissue procurement for transplantation in Poland (1990-95).

From 1963 Professor Ostrowski has collaborated with the Polish Academy of Sciences. He was a vice-secretary of the Medical Division (1965-72), chairman of the Transplantology Commission (1963-76), and member of many committees, councils, scientific associations and editorial boards.

Professor Ostrowski was elected as a vice-president of European Cell Biology Organisation (1964-65), he was a member of the Board of the Transplantation Society (1977-82), an expert for the World Health Organisation (1978-79). He was visiting professor at the Universities of Cambridge (1972) and Sassari (2007). He established close cooperation with international institutions including the Institute of Biology of Human Reproduction in Lyon, Fibiger Laboratory in Kopenhagen, Wistar Institute in Philadelphia and the Enzymology Unit in Buffalo.

Professor Ostrowski created a school of theoretical medicine, promoting 30 doctors of medical sciences, 17 of his coworkers received habilitation, 21 became professors, four of them in the US and one in Great Britain. He published about 300 papers and eight handbooks. His remarkable scientific work led to an Honorary Doctorate of the University of Orleans (1981), the Warsaw Medical University (2007), and he was elected a member of the Polish Academy of Sciences and Arts in Krakow. He was granted Honorary Membership by the Polish Association Histochemists and Cytochemists, Polish Transplantation Society, European Association of Tissue Banks, Society of Hungarians Anatomists, Histologists and Embriologists and Geselschaft fur Histochemie. He received state prizes from the Polish Academy of Sciences (1982), Minister of Science (1985), Minister of Health and Welfare (1988) and the Prime Minister (2003) and was decorated with Gold's Cross of Merit (1954), Order of Polonia Restituta, Knight's Cross (1969), Officer's Cross (1984) and the Commandor's Cross (2000).

Professor Kazimierz Ostrowski was recognized as a very creative scientist with enormous energy and imagination. He will be remembered as a keen initiator in many research fields. He was very kind and friendly to his coworkers, who mourn his death.

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# Obituary: Professor Rudolf Klen, MD, DrSc (1915-2006)

Professor Rudolf Klen, M.D., DrSc., was one of the outstanding personalities, scientists and founders of science - based tissue banking in Europe. He was born on 2nd July 1915 in Prague to a Jewish family. He attended primary and secondary schools in a small Czech city Kolin, some 50 miles from Prague and started his medical studies in 1934 at the Charles´ University in Prague. He was due to graduate when the 2nd World War broke out. During the German occupation of Czechoslovakia all Czech Universities were closed and he could not complete his studies. Due to his Jewish origin he was persecuted by the Nazis, subsequently captured and kept for almost four years in Nazi concentration camps including Terezin, Auschwitz, Kaufering, Landshut and Dachau. By some miracle he survived Nazi torture and after the war finished, he completed his medical studies and graduated as a medical doctor from Charles University in 1946.

Freshly graduated Doctor Klen started his medical career at the Department of Public Health of the Bohemian Government in Prague. Later he worked at the Department of Forensic Medicine in Hradec Kralove, a district city 80 miles north-east of Prague. Around this time, he read about the establishment of the New York Eye Bank and the US Navy Bone Bank in the USA and was inspired to set up a similar institution in Czechoslovakia. He was working in an institution where potential tissue donors have always been available and in 1951 he developed the basic concept for establishing a tissue bank in Hradec Kralove and submitted it to the hospital authorities and public discussion. The result was that on October 1st 1952 the first European tissue laboratory began its activity under his leadership and over the next few years, the Hradec Kralove Tissue Laboratory (HKTL) prepared various kinds of tissue grafts for transplantation purposes and supplied hospitals throughout Czechoslovakia.

Over the following years HKTL gradually started international collaboration, particularly with the WHO, resulting in delivery of tissue grafts to many foreign institutions – 25 institutions across four continents. In 1957 his first monograph "Tissue Banking" was published by Avicenum in Czech. In 1962 the monograph was translated into Russian and in 1982 a new edition called "Biological Principles of Tissue Banking" was published by Pergamon Press, Oxford. This monograph represented one of the very first comprehensive scientific publications in the field of tissue banking. In 1963 Dr. Klen defended his thesis and received his scientific degree "CSc." (the title is equal to current PhD). In 1966 he was promoted to Associate Professor, and in 1969 he received the scientific degree of Doctor of Medical Sciences (DrSc). Unfortunately, Dr. Klen was a "persona non grata" for the Communist Government of Czechoslovakia as he did not agree with the Russian occupation of the country, following the Prague Spring of 1968. In spite of his pedagogical, scientific and publishing activities, including achieving the highest scientific degree – DrSc., for political reasons he had to wait another 23 years to receive his full professorship of the Charles University in 1992. In the 80's he was actively collaborated with the International Atomic Energy Agency (IAEA) as an expert and assisted tissue bank establishment in Rangoon (former Burma, now Myanmar) and Calcutta, India. Many tissue bank scientists and technicians from developing countries visited under fellowships to the Hradec Kralove Tissue Laboratory under the IAEA program.

Professor Klen was founder and member of several scientific societies, such as the Czechoslovak Biological Society (Honorary Member since 1987), Indian Cryogenic Council (Honorary Member since 1987), and European Association of Tissue Banks (EATB, first Honorary Member since 2001). His scientific publications included four monographs (some of them translated to several foreign languages), and 167 scientific papers published both in Czechoslovakian scientific journals and in renowned international journals. Professor Klen's activities covered almost the whole field of tissue banking, from legal and ethical issues, to methods of harvesting, decontamination, and banking of osteochondral tissues, cornea, dura mater, and skin. He was also active in organ transplantation, primarily in kidney preservation. He instituted broadly-based, fruitful collaboration with clinical specialists at University Hospital and was respected by tissue bankers worldwide.

Professor Klen was lucky to live to receive scientific and social recognition, even though this came late. In 1996 Professor Klen was awarded Honorary Membership of the Czech Transplant Society. The Board of the European Association of Tissue Banks, considering his huge contribution to development of tissue banking in the European region and worldwide, launched the EATB Rudolf Klen award for recognition of science, development and progress in tissue banking in Europe. The first Rudolf Klen award was conferred in 2007 to Jan Koller.

Despite his advanced age he was scientifically active as Advisor to the Transplant Center in Hradec Kralove almost until he died on 24th October 2006 at the age of 91 years.

**Assoc. Prof, Jan Koller, M.D., PhD**
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# Next Edition

**Issue 33 will feature articles on:**

- Cell Salvage Update
- Bacterial Screening of Platelets
- AIR Study – Antibodies in Pregnancy
- Coagulation Update
- Overview of the Joint NHSBT/HPA Epidemiology Team
- Management of Trauma in a London Teaching Hospital
- The Tissue Services Retrieval Team
- The Nuffield Council's Current Work on Human Bodies in Medicine and Research

If you would like to comment on any of the articles in this edition of ***Blood and Transplant Matters*** please email the Editor: derwood.pamphilon@nhsbt.nhs.uk

## CPD Questions

**BCSH Guidelines for the Administration of Blood Components**

**1. The latest guidelines for the above were last reviewed and updated in:**

A. 1996.

B. 1999.

C. 2006.

D. 2009.

**2. In the latest guidelines for the above**

A. Management of transfusion reactions is discussed.

B. Clearly states only a doctor can prescribe blood components.

C. Recommend an individual receives training and/or competency assessment at least every three years.

D. Observation at 15 minutes now includes blood pressure.

**3. Component Development in NHSBT**

A. All work is kept internally and not published.

B. Most of the work is carried out in the Brentwood Centre.

C. Novel components are assessed only by extensive laboratory studies.

D. This is performed outside the guidance of the Red Book.

**4. Use of prion reduction filters**

A. Causes a loss of 5-7g of haemoglobin from red cell concentrate.

B. Significantly affects the quality of the red cell concentrate.

C. Is impossible to quality monitor.

D. Does not affect residual plasma left in the red cell concentrate.

**5. Finding Compatible Blood for Patients with Difficult Antibodies**

A. All referrals to IBGRL are for patients with rare blood groups.

B. Patients with rare blood groups do not form antibodies to common specificities.

C. Extended red cell phenotyping is not recommended.

D. Transfusion dependant patients with sickle cell disease can present major difficulties for the provision of compatible blood.

**6. Optimisation of haemoglobin prior to elective surgery**

A. Most anaemia detected pre-operatively is not due to iron deficiency.

B. Ideally anaemia should be detected prior to a surgical referral.

C. Detection of anaemia pre-operatively does not require an investigation of the cause.

D. Managing anaemia pre-operatively does not reduce transfusion rates.

**Platelet Transfusion Thresholds**

**7. Adults and Children**

A. Gross haemorrhage rarely occurs at platelet count above $20x10^9$/L.

B. A platelet count of $20x10^9$/L was observed to be a 'threshold' for bleeding.

C. Platelet Transfusion Trigger Trials confirmed the $20x10^9$/L platelet count threshold.

D. All platelets are given for therapeutic purposes only.

**8. Neonates**

A. A platelet count of $50x10^9$/L is a well established threshold.

B. A platelet count of $25x10^9$/L is a well established threshold.

C. Small, preterm babies do not require a higher threshold.

D. Best opinion at present is that term infants are unlikely to bleed if the platelet count is maintained above $20x10^9$/L.

**Cell Therapy for Articular Cartilage Repair**

**9. Autologous Chondrocyte Implementation (ACI)**

A. Post-traumatic osteoarthritis affects the knee much more than the ankle.

B. ACI has up to 16 years follow-up and more than 80% of treated patients show sustained improvement.

C. ACI is a very cheap treatment.

D. There exists conclusive evidence that ACI is clinically superior to other treatments.

**10. Stem Cells as Chondrocyte Substitutes**

A. Articular Chondrocytes are easy to expand.

B. Mesenchymal stem cells are difficult to isolate and expand.

C. Mesenchymal stem cells are Chondrogenic.

D. Mesenchymal stem cells are in routine use in ACI procedures.

**A Day in the life of a Scientist in a Regional Burns Centre**

**11. The Regional Burns Centre based at University Hospitals Birmingham NHS Foundation Trust**

A. Uses over 75,000 $cm^2$ of cryopreserved skin each year.

B. Provides all the cryopreserved allograft skin.

C. Is exempt from the Human Tissue Authority.

D. Only processes and stores cryopreserved allograft skin.

**12. Bone Marrow Derived Cell Preparations for Cardiac Repair in Patients with Coronary Heart Disease**

A. Heart failure is diminishing in the UK.

B. The diagnosis of heart failure carries a one year mortality of approximately 40%.

C. Recent studies have suggested that bone marrow-derived $CD34^+$ $CXCR4^+$ progenitor cells produced a significant improvement in patients with acute STEMI involving the left anterior descending artery.

D. Intracoronary injection of unselected bone marrow mononuclear cells into infarcted regions at the time of CABG produced an increase in ventricular function.

**13. Bone Marrow Derived Cell Preparations for Cardiac Repair in Patients with Coronary Heart Disease**

A. Bone Marrow derived cell preparations trans-differentiate into a cardiac myocyte phenotype with high efficiency.

B. Cytokine mobilisation of haemopoietic progenitor cell preparations has proved effective in patients with acute STEMI.

C. Administration of these cells by a number of routes appears safe.

D. Additional clinical trials, rather than pre-clinical work, are required.

**Knowledge is Power: The ABC Experience**

**14. America's Blood Centers (ABC)**

A. Only provide blood to the United States of America.

B. Only started a newsletter in the 1980's.

C. Keep information and knowledge internally.

D. Have adopted the UK Blood Stocks Management Scheme.

**The World Health Assembly Resolution on Organ and Tissue Transplantation**

**15. World Health Organisation Guiding Principles on Human Organ Transplantation**

A. The first Guideline Principles were endorsed in 1991.

B. The revised Guideline Principles were completed in 2005.

C. The revised Guideline Principles were endorsed in 2009.

D. The revised Guideline Principles are the same as the first.

# Diary Dates

## 2011

**27-29 January 2011**

**T-cell Lymphoma Forum 2011.**

**Location:** Hotel Nikko, San Francisco, CA
For more information contact Damaris Cruz on 201-594-0400 or dcruz@jwoodassoc.com
You can view the programme and register online: http://www.tclf2011.com
**Details:**
This forum will provide a platform for discussion about the classification, epidemiology, prognosis, and pathogenesis of several T-cell lymphoma subtypes. In addition, the latest information on novel agents and treatment approaches will be presented by T-cell lymphoma experts. This meeting is intended for haematologists, oncologists, and other clinicians and scientists with an interest in T-cell lymphoma.

**10 March 2011**

**Identifying T Cell Subset Phenotype and Function in Infections.**

**Location:** BioPark Hertfordshire, Broadwater Road, Welwyn Garden City, Hertfordshire AL7 3AX, UK
For more information contact: Astrid Englezou tel: 08714 890 134 or via email enquiries@euroscicon.com
The programme can be viewed and to register online go to: http://www.regonline.co.uk/tparasite09

**25-27 March 2011**

**An Update on the Management of Haematological Malignancies (ESH International Conference).**

**Location:** Cairo, Egypt
For more information contact:
http://www.esh.org

**1-2 April 2011**

**Haemoglobin Disorders: Laboratory Diagnosis and Clinical Management (ESH-Enerca Training Course).**

**Location:** Brussels, Belgium
For more information contact:
http://www.esh.org

**3-6 April 2011**

**EBMT 2011 Congress.**

**Location:** Le Palais de Congrès, Paris, France
For more information contact:
http://www.congrex.ch/ebmt2011 or
http://www.ebmt.org

**11-15 April 2011**

**Manchester Blood Coagulation Course.**

**Location:** Manchester City Centre, UK
For more information contact Jan Dixon on jan.dixon@uhsm.nhs.uk or Tel: 0161 291 4767
**Details:**
This well-established course is intended to prepare candidates for the Royal College of Pathologists examination in blood coagulation. It is held in city centre Manchester at the Manchester Cathedral Visitor Centre.

**14-16 April 2011**

**Blood Group Serology, Reading (Conference).**

**Location:** Reading University, UK
For more information contact:
http://www.bgsreading.org

**29 April-1 May 2011**

**Cancer Stem Cells (ESH International Conference).**

**Location:** Mandelieu, France
For more information contact:
http://www.esh.org

**9-11 May 2011**

**Blood and Marrow Transplantation (ESH-EBMT Training Course).**

**Location:** La Baule, France
(80km from Nantes on the Atlantic Coast)
For more information contact:
http://www.esh.org

**18-21 May 2011**

**11th International Symposium on Myelodysplastic Syndromes (MDS 2011).**

**Location:** Edinburgh

For more information contact
Dina Davis on ddavis@kenes.com
You can view the programme and register online:
http://www.kenes.com/mds

**Details:**

The MDS represents an important meeting point for haematologists and MDS experts from all over the world to promote the ongoing exchange of information relating to MDS and is established as the leading forum for the exchange of knowledge in this field.

MDS 2011 will feature a rich scientific program, focusing on such topics as immunopathogenesis, molecular pathogenesis, molecular mechanisms and drug targets, new therapies and transplantation.

**6 July 2011**

**SHOT Annual Symposium 2011.**

**Location:** The Royal Society of Medicine,
1 Wimpole Street, London, UK
For more information contact the SHOT Office:
shot@nhsbt.nhs.uk or Tel: 0161 423 4208

**22-25 September 2011**

**Chronic Myeloid Leukaemia – Biological Basis of Therapy (ESH-ICMLF International Conference).**

**Location:** Estoril, Portugal
For more information contact:
http://www.esh.org

**14-16 October 2011**

**Acute Myeloid Leukaemia – 'Molecular' (ESH-EHA Scientific Workshop).**

**Location:** Mandelieu, France
For more information contact:
http://www.esh.org

**22-25 October 2011**

**AABB Annual Meeting and CTTXPO 2011.**

**Location:** San Diego, California, USA
For more information contact:
http://www.aabb.org/events

**Details:**

Learn the latest in blood banking, transfusion medicine and cellular and related biological therapies.

**27-30 October 2011**

**Cord Blood Transplantation and Immunobiology of Haematopoietic Stem Cell Transplant (ESH International Conference).**

**Location:** Rome, Italy
For more information contact:
http://www.esh.org

**7-10 November 2011**

**Thrombosis and Hemostasis (ESH-ISTH Advanced Course).**

**Location:** Cascais, Portugal
For more information contact:
http://www.esh.org

**20-23 November 2011**

**XXIInd International Congress of the ISBT, Asia.**

**Location:** Taipei, Taiwan
For more information contact:
http://www.isbt-web.org

**A full diary of events and training courses can be viewed on the following websites:**

**www.transfusionguidelines.org.uk**
**www.blood.co.uk/hospitals**
**www.bbts.org.uk**

## Notes

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Blood and Transplant Matters is prepared
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