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Improved Iron Chelation Therapy: Implications for Transfusion Medicine in Developing Nations

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**Background:** The Maldivian Islands have one of the highest incidences of β thalassemia in the world. Treatment of β thalassemia is characterized by two distinct phases: treatment of nature's disease (anemia) and the secondary treatment of a side effect (transfusional iron overload) of our cure. Current iron chelation therapy using Desferal (DFO) is problematic due to its short vascular half-life, frequency of injections, toxicity and expense. To address this problem, we have developed new high molecular weight (HMW) iron chelators exhibiting reduced toxicity and prolonged vascular retention.

**Methods:** To test the utility of these chelators, in vitro studies using iron loaded HepG2 liver cells (0-500 µM Fe3+; ferric ammonium citrate, FAC; 48 hours) were done. Iron chelation studies utilized either single or combinational treatment with 0-200 µM L1 or DFO (low molecular weight chelator) and/or S-DFO (a HMW derivative of Desferal) for 0-48 hrs. The efficacy of treatment was assessed by cellular ferritin, Perl's iron stain, transmission electron microscopy (TEM) and cell viability assays.

**Results:** Iron treatment alone resulted in a significant increase in intracellular ferritin and histochemical iron staining. Treatment with either L1 or S-DFO alone demonstrated modestly decreased ferritin levels and iron staining. Importantly, combination therapy (L1+S-DFO), resulted in a synergistic effect resulting in a 79% decrease in FAC-driven ferritin levels after 48 hours. TEM studies of FAC treated, but not control, cells demonstrated iron accumulation and organellar and structural changes with electron dense iron deposits. Chelator treatment of these cells reversed these ultra-structural changes.

**Conclusion:** The development of improved HMW chelators may provide better therapeutic value (less toxic and less frequent administration) in developing nations. Consequent to this, iron mediated pathology to patients would be diminished resulting in less cost to already strained public health budgets.

**Keyword(s):**
- iron
- thalassemia
- iron chelation
- transfusion

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**Notes**

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Pathogen reduction technologies (PRT) dramatically reduce the risk of infectious contamination of blood components. However, several in vitro studies have revealed a negative impact on the quality of platelet concentrates after PRT treatment which is expressed as a general acceleration of the platelet storage lesion. In order to minimize this undesired side effect, it is necessary to understand the underlying molecular mechanisms triggered by the PRT treatment. We have recently applied a proteomic approach to analyze alterations in the platelet proteome after MIRASOL (riboflavin/UV) treatment revealing an acceleration of the actin rearrangement during storage compared to the control without irradiation. Here we present the analyses of signal transduction in platelet concentrates during MIRASOL treatment targeting the phosphorylation levels of protein kinases using an antibody array. Among several candidates, p38 MAPK showed a 3.2-fold increase in phosphorylation, hence activation, during the PRT procedure. In this study two buffy coat platelet concentrates were pooled and split into four identical units. One was spiked with 10 μM SB203580, a p38 MAPK-specific inhibitor, one was spiked with 10 μM LY294002, a PI3-kinase-specific inhibitor shown in our previous studies to decelerate platelet storage lesion, one containing only the inhibitor solvent and the last one as a not irradiated control. Sample analyses were carried out immediately after irradiation as well as on day 2, 5 and 7 during storage. The unit with the LY294002 inhibitor showed similar or slightly improved platelet quality compared to the irradiated unit; however, the unit containing the SB203580 inhibitor revealed a 33% decrease in platelet activation (p<0.01) as determined by the expression of P-selectin on the platelet surface, a 26% higher glucose level (p<0.1) and a subsequent 20% decrease in lactate accumulation (p<0.01) at day 7 of storage compared to the irradiated control platelet unit containing only the inhibitor solvent. Although, this inhibitor cannot be used in platelets for transfusion purposes, these findings provides an important insight into understanding the insult triggered by the PRT treatment and identify a potential targets for intervention.

Keyword(s):
- platelet concentrate
- platelet in vitro quality
- pathogen reduction technology
- p38 MAPK inhibitor
Retention of Acceptable Coagulation Factor Content and Increase in DEHP in Frozen Plasma (FP) Thawed and Refrigerated for Five Days

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Thawed plasma is typically transfused to supply coagulation factors to patients but clotting factor activity declines during storage. In 2010, CAN/CSA Z902-10 extended the maximum refrigerated storage time prior to transfusion of thawed FP from 24 to 120 hours.

Purpose: To determine the effects of extended storage on coagulation factor and di(2-ethylhexyl) phthalate (DEHP) plasticizer levels in FP.

Methods: FP units prepared using B1 (“top / bottom”) collection sets were thawed, refrigerated, and sampled aseptically at 0, 24, 72, and 120 hours post-thaw (n=54). Clotting factor activities and prothrombin times (PT) were measured using an automated coagulation factor analyzer. DEHP was measured by HPLC after hexane extraction (n=11). Unit sterility was confirmed using the BacT/ALERT system.

Results: FV and FVIII declined significantly within 24 hours, while FVII losses did not become significant until 120 hours. By that time, average losses were 20, 14, and 41%, in FV, FVII, and FVIII, respectively; fibrinogen activity did not change. PT values were elevated by 9% at 120 hours. Mean DEHP levels rose from 22 ppm at thaw to 66 ppm at 120 hours. Factor levels in thawed FP remained well above levels needed to support surgical hemostasis (> 0.30 IU/mL).

Conclusions: FP units thawed and refrigerated for 120 hours contain acceptable levels of clotting factors, and their DEHP levels are not of concern for adult patients. For neonatal / pediatric patients, DEHP levels can be kept as low as reasonably achievable by using FP refrigerated for no more than 24 hours.

Keyword(s):
- plasma
- storage
- coagulation
- DEHP
Induction of Immunotolerance via Polymer Grafting to Allogeneic Donor Blood Cells

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**Background:** Alloimmunization to donor RBC is a significant problem in chronically transfused patients. Our previous in vivo murine studies of immunocamouflaged RBC demonstrated reduced immunogenicity of the methoxy(poly(ethylene glycol))-modified RBC (mPEG-RBC). We subsequently hypothesized that the immunocamouflage of donor leukocytes would result in the enhanced induction of tolerance and/or anergy. This therapeutic intervention could be used to reduce the risk of rejection of allogeneic cells or tissues.

**Methods:** To test this hypothesis, control or mPEG-modified syngeneic or allogeneic splenocytes were injected (i.v.) into H-2 disparate mice. The effects of the control and modified donor cells on the murine immune system was assessed by changes in T regulatory (Treg) and inflammatory (Th17) T lymphocytes in the peripheral blood, spleen and brachial lymph node of the recipient animals. Levels of Treg and Th17 lymphocytes were assessed at 1-5 and 30 days post challenge. In some mice, a secondary challenge with unmodified allogeneic cells was done at day 25 and Treg and Th17 cells were assessed 5 days post challenge.

**Results:** PEGylated allogeneic splenocytes resulted in a significant in vivo immunosuppressive effect as evidenced by increased Treg and decreased Th17 cells (p<0.001 for both; ≥ 2 days) in the spleen, lymph nodes and blood. In contrast, unmodified allogeneic splenocytes resulted in dramatically increased levels of Th17 and decreased levels of Treg cells (p<0.001 at ≥ 2 days). Secondary challenge of tolerized, but not control, mice with unmodified allogeneic cells demonstrated almost complete attenuation of the expected inflammatory response (e.g., no increase in Th17 cells).

**Conclusions:** The PEGylation of donor splenocytes yields a tolerogenic state in the recipient mice as measured by both in vitro and in vivo studies. This tolerogenic state was stable (>30 days) and prevented subsequent immune responses to later challenges with allogeneic cells.

**Keyword(s):**
- PEGylation
- transfusion
- immune tolerance
- treg/Th17 lymphocytes
Purpose: In May 2010, Canadian Blood Services (CBS) began testing ‘at risk’ donors for Chagas Disease. This paper describes the results of donor testing, follow up and lookback investigations after six months of donor testing.

Methods: All donors who answer yes to any of three risk questions (birth or maternal birth in an endemic area, or having spent >6 months in an endemic area) are tested for Chagas antibody using Abbott PRISM Chagas, an automated chemiluminescent assay. Repeat reactives (RR) are confirmed at the National Parasitology Reference Lab at McGill University. Confirmatory testing includes ELISA, immunoblot and PCR. Samples are sent to the U. S. Quest Diagnostics Lab for RIPA testing. Lookback is performed on every confirmed positive donor.

Results: To date, 8 RR donors have been identified out of 7,290 donors tested (approximately 1.3% of donor population), and 7 have confirmed positive. Six male and one female donor are from Manitoba (3), Ontario (2) and B.C. (2) and range in age from 25 to 63 years. Five donors were born in Paraguay, 1 in Argentina, 1 in Canada. Five donors had lived in rural areas, four had mothers, and 3 had grandmothers born in an endemic country. Lookback identified 165 products transfused. Forty five living recipients were notified and 24 recipients have tested negative to date.

Conclusions: The CBS Chagas Donor screening program has identified seven confirmed positive donors who were unaware that they had been infected.

Keyword(s):
• Chagas disease
• donor testing
• lookback

Notes
Enhancing the Homing-Related Responses of Cord Blood Stem Cells by Hyaluronic Acid and Thrombin

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Umbilical cord blood (CB) is increasingly used as a source of hematopoietic stem/progenitor cells (HSPC) for transplantation; however, delayed engraftment is a major complication of this therapy. CB transplantation in adult patients is restricted by the limited number of HSPC available in a single CB unit. Because HSPC collected by leukapheresis products, namely hyaluronic acid (HA) and thrombin, have the ability to enhance the homing-related responses of CB HSPC. Specifically, we investigated the effects of HA and thrombin on the activity of membrane type 1-matrix metalloproteinase (MT1-MMP), an extracellular matrix-degrading enzyme localized at the leading edge of migrating cells. CB CD34+ HSPC were examined for MT1-MMP expression by RT-PCR, Western blot, and confocal microscopy for MMP-2 activation by zymography, and for transmigration by reconstituted basement membrane (Matrigel) assay. We found that HA and thrombin increase i) the transcription and protein synthesis of MT1-MMP; ii) the levels of active MMP-2 in cultures with BM stromal cells' and iii) trans-Matrigel migration towards a low gradient of the chemokine stromal cell-derived factor-1, which was significantly decreased by silencing with MT1-MMP siRNA. Thus, our data suggest that MT1-MMP plays an important role in the homing-related responses of HSPC, and we propose that pretreatment of CB HSPC with HA or thrombin before transplantation could improve their homing and hasten their engraftment.

Keyword(s):
• cord blood
• stem cell transplantation
• thrombin
• hyaluronic acid

Notes
Expanded Quality Monitoring of CBS Frozen Plasma: Comparison to Solvent-Detergent Plasma

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Canadian Blood Services R & D, Ottawa and Hamilton, ON, and Vancouver, BC

Frozen plasma (FP) for transfusion in Canada (excepting Quebec) is produced by the buffy coat method of processing the whole blood donation of a single donor, and frozen within 24 hours of phlebotomy. Solvent-detergent (S/D) plasma (e.g. Octaplas; Octapharma) is a pathogen-reduced, pooled plasma product with potential safety and consistency advantages over FP; however, the S/D manufacturing process has been reported to lead to losses of specific plasma proteins such as protein S (PS) and alpha-2-anti-plasmin (AP). We expanded our annual quality monitoring survey of CBS FP to include these factors.

**Purpose:** To compare the clotting activities of CBS FP to published Octaplas data.

**Methods:** 131 FP units were tested for the parameters listed below by either automated or manual means.

**Results:** Means and 95% confidence intervals were determined and are shown in tabular form (Octaplas data from Transfusion Med 2007;17:318-20).

<table>
<thead>
<tr>
<th>Test</th>
<th>CBS FP (n=131)</th>
<th>Octaplas (n=12)</th>
</tr>
</thead>
<tbody>
<tr>
<td>APTT (sec)</td>
<td>37.8 (37.1-38.6)</td>
<td>35.2 (34.7-35.7)</td>
</tr>
<tr>
<td>PT (sec)</td>
<td>13.4 (13.2-13.5)</td>
<td>13.2 (13.1-13.4)</td>
</tr>
<tr>
<td>FV (IU/ml)</td>
<td>0.87 (0.84-0.90)</td>
<td>0.78 (0.77-0.80)</td>
</tr>
<tr>
<td>FVII (IU/ml)</td>
<td>0.98 (0.94-1.02)</td>
<td>1.08 (1.05-1.12)</td>
</tr>
<tr>
<td>FVIII (IU/ml)</td>
<td>0.85 (0.70-0.90)</td>
<td>0.68 (0.63-0.73)</td>
</tr>
<tr>
<td>FX (IU/ml)</td>
<td>0.94 (0.93-0.95)</td>
<td>0.78 (0.77-0.79)</td>
</tr>
<tr>
<td>PS (IU/ml)</td>
<td>0.97 (0.94-1.00)</td>
<td>0.64 (0.61-0.66)</td>
</tr>
<tr>
<td>AP (IU/ml)</td>
<td>0.91 (0.89-0.92)</td>
<td>0.23 (0.22-0.25)</td>
</tr>
<tr>
<td>FGN (g/l)</td>
<td>2.94 (2.83-3.05)</td>
<td>2.49 (2.46-2.83)</td>
</tr>
</tbody>
</table>

**Conclusions:** On average, FP contains not only more PS and AP activity than Octaplas, but also more FV, FVIII, FX, and fibrinogen than the S/D plasma product. S/D plasma products may not be appropriate substitutes for FP for coagulopathic indications.

**Keyword(s):**
- plasma
- quality
- octaplas
- coagulation

Notes

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8 - CSTM 2011

CONFERENCE ABSTRACTS
Amelioration of Murine Immune Thrombocytopenia by Monoclonal Antibodies: A Potential Therapy for ITP?

Andrew R Crow*, Seng Song, Sara J Suppa, and Alan H Lazarus
Canadian Blood Services and St. Michael’s

Intravenous immunoglobulin (IVIg) is used to treat autoimmune diseases such as ITP. IVIg is a limited resource and its dosage and cost are both high. Although considered safe, it will always carry a theoretical risk of transferring infectious disease. Thus it would be highly desirable to improve the efficacy of IVIg or develop monoclonal antibodies, capable of mimicking the clinical effects of IVIg. To explore the potential for monoclonal antibodies as a treatment for ITP as well as further explore their mechanisms of action, we tested 8 monoclonal antibodies in murine ITP and found 4 antibodies which could successfully ameliorate ITP. The antibodies employed were all specific for the CD44 antigen. Two of these antibodies function at a full 3-log fold lower dosage as compared to IVIg. Further characterization of the 2 most successful antibodies demonstrated that, similar to IVIg, i) the presence of the inhibitory IgG receptor FcγRIIB was required for their ameliorative function, ii) complement deficient mice responded to anti-CD44 treatment, and iii) human transgenic FcγRIIA expressing mice also responded to the CD44 therapeutic modality. Dissimilar to IVIg, the Fc portion of the CD44 antibody was not required. These data demonstrate that CD44 antibodies can function therapeutically in murine ITP and that they could potentially provide a very low dose recombinant therapy for the amelioration of human ITP.

Keyword(s):
- ITP
- IVIg replacements
- monoclonal antibody
- IVIg
The Use of Antithrombin III for Heparin Resistance in Patients on Cardio-Pulmonary Bypass

Sholzberg M*, Callum J, Lin Y

**Background:** Antithrombin III (ATIII) concentrate is currently being used to treat heparin resistance (HR) in patients on cardio-pulmonary bypass (CPB) to achieve a therapeutic activated clotting time (ACT) and avoid thromboembolic events. There is little known about appropriate dosage, risks and benefits of ATIII in the setting of cardiac surgery. Furthermore, there is no consensus on risk factors associated with the development of ATIII dependent and independent HR. Purpose: To collect safety and effectiveness information on the use of 1000 units (U) of ATIII for HR in patients on CPB. Also, to identify risk factors associated with ATIII dependent and independent HR.

**Methods:** Data were obtained via retrospective chart review of all patients who were treated with ATIII for HR, from 2003 to 2010 at our institution.

**Results:** Between 2003-2010, 15 patients were treated with 1000 U of ATIII for HR. The mean age was 58 years and 80% of patients were male. 53% of patients were obese and 33% were overweight according to their calculated body mass index (BMI). After infusion of ATIII, 100% of patients obtained an ACT above 480 seconds. None of the patients required re-operation. 2 patients (13%) experienced thromboembolic events (internal jugular vein thrombosis and pulmonary embolism) perioperatively.

**Conclusion:** We observed a correlation between BMI and HR that does not represent underdosing of intraoperative heparin. All patients who received 1000 U of ATIII had the desired effect of raising the ACT. Neither hemorrhagic nor thromboembolic risk seemed to be elevated with this dose. These findings support the safety of ATIII at 1000 U for HR to achieve a higher ACT but due to sample size we cannot conclude definitively on its clinical effectiveness.

**Keyword(s):**
- antithrombin
- heparin resistance
- cardiopulmonary bypass
- cardiac surgery

**Notes**
Background: The need for red blood cell transfusion is common in patients with myelodysplastic syndrome (MDS). There are few reports on the rate of alloimmunization in MDS patients but it has been reported to be as high as 32%.

Purpose: To minimize the rate of alloimmunization, our institution adopted a policy of prophylactic Rh and Kell (K) blood group matching for transfusion-dependent patients with MDS in 2007. The purpose of this study was to determine 1) the rate of alloimmunization in our MDS patients and 2) how frequently the policy was followed.

Methods: A retrospective review of the MDS database was conducted.

Results: As of June 24, 2010, there were a total of 270 patients in the MDS database; of these, 80 patients were transfused at our institution. The mean age of the patients was 73 years, with 39% female patients. 35 of the 80 (44%) patients had documented Rh/K matching. 35 of the 80 transfused MDS (44%) patients had documented Rh/K matching. Alloantibodies were detected in 12 of the 80 (15%) patients: 10 prior and 2 after Rh/K matching. Of the 10 patients with alloantibodies prior to Rh/K matching, 3 had allo-antibodies prior to being transfused for MDS. Of the remaining 7, 4 patients had anti-E or anti-K that could have been prevented had Rh/K matching been instituted prior to transfusion.

Conclusion: We observed an alloimmunization rate of 15% in this patient population. With full implementation of Rh/K prophylactic matching, the rate could have been decreased to 10%. This lends support to the policy of prophylactic Rh/K matching for MDS patients.

Keyword(s):
- MDS
- alloimmunization
- prophylactic
- Rh

Notes
Trigger HB Values for Transfusion Dependent Anemias in a Regional Transfusion Service

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Purpose: The number of patients who require chronic transfusion support is increasing in our practice. We reviewed data of patients defined as receiving more than two units of red cells per month for a period of at least three months to determine the trigger hemoglobin (Hb) values for categories of recipients.

Methods: Using the lab information system, an audit of transfusion dependent recipients between January and June of 2008 was performed. Primary diagnosis, indication for transfusion, total units per episode and the pre / post transfusion Hb (48 hours) values were obtained.

Results: A total of 3,335 red cell transfusion episodes were retrieved. The mean trigger Hb was 79.8g/L (±36.9) while the post transfusion Hb was 98.0g/L (±12.4) but variability was clear on basis of diagnosis.

Conclusion: Transfusion dependent surgical oncology patients have a higher mean trigger Hb value while myelofibrosis patients have the lowest mean trigger Hb for transfusion. The optimal trigger Hb values for transfusion dependent patients depend on the diagnosis and may also be physician dependent.

<table>
<thead>
<tr>
<th>Primary Diagnosis</th>
<th>Mean Pre-tx Hb ±SD</th>
<th>No of Units transfused</th>
<th>Mean Post-tx Hb ±SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Myelodysplastic syndrome</td>
<td>80.5(±64.1)</td>
<td>862</td>
<td>98.5(±10.7)</td>
</tr>
<tr>
<td>Solid tumors (non-Hematological)</td>
<td>85.5(±10.2)</td>
<td>218</td>
<td>106.2(±10.7)</td>
</tr>
<tr>
<td>Lymphoma</td>
<td>78.7(±6.8)</td>
<td>32</td>
<td>93.9(±9.9)</td>
</tr>
<tr>
<td>Pure red cell aplasia</td>
<td>79.2(±11.6)</td>
<td>103</td>
<td>98.6(±6.9)</td>
</tr>
<tr>
<td>Myelofibrosis</td>
<td>72.3(±9.9)</td>
<td>131</td>
<td>86.9(±9.2)</td>
</tr>
<tr>
<td>Aplastic anemia</td>
<td>74.8(±6.1)</td>
<td>96</td>
<td>94.1(±8.3)</td>
</tr>
<tr>
<td>Anemia of Chronic disease</td>
<td>76.5(±6.3)</td>
<td>42</td>
<td>101.3(±9.4)</td>
</tr>
</tbody>
</table>

Keyword(s):
- chronic transfusion
- hemoglobin triggers

Notes
Hypothremic Storage Decreases Erythrocyte Deformability Independent of ATP Levels

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Red blood cell (RBC) hypothemic storage lesion describes an array of biochemical and biomechanical events that result in deteriorating RBC quality during the 42-day ex vivo storage, and is associated with detrimental clinical outcomes. This study examines the effects of hypothemic storage (HS) on RBC deformability. The effect of HS on RBC in vitro quality was assessed on leukoreduced CPD-SAGM packed RBC units (n=6) by several assays, including % hemolysis (Drabkin’s), ATP concentration (spetrophotometry), RBC indices (Coulter) and deformability (ektacytometry) weekly for a total of 49 days. After 6 weeks of HS, RBCs were rejuvenated with a PIPA solution. Results were statistically analyzed using Mann-Whitney U test. HS resulted in significant decrease in RBC deformability after 5 weeks of storage (p=0.001). Hemolysis increased throughout HS with statistically significant differences at day 15 (p = 0.008), but it remained bellow 0.8 %. ATP levels decreased significantly to 1.9 ± 0.2 µmol/gHb after 7 weeks of HS (p = 0.006). PIPA rejuvenation resulted in significant increases in RBC ATP levels (p=0.002), but it failed to improve RBC deformability, as rejuvenated RBCs were less deformable than control RBCs (Emax=0.503 and 0.600, respectively; p = 0.002). Results of this study demonstrate significant decrease in the deformability of stored RBCs, which precedes ATP depletion and is not reversed by ATP rejuvenation. New strategies are needed to combat the biomechanical component of the RBC storage lesion.
Oral Clinical

Consequences of Plasma Incompatible Platelet Transfusions

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Transfusion of plasma incompatible platelet concentrate may result in hemolytic transfusion reaction. The incidence of this complication has been estimated at 1 in 9,000. The purpose of our study was to determine the incidence of positive direct antiglobulin test (DAT), a decrease in Hb and biochemical evidence of hemolysis after transfusion of blood group O platelets to non-O recipients. Methods: All patients receiving plasma incompatible platelets over a period of Dec 1, 2009 to Nov 30, 2010 had DAT tested on the subsequent blood bank sample received. If DAT was positive, an eluate was performed to identify the antibody coating patient RBC. From the electronic records, we ascertained the lowest Hb within 72 hours post-transfusion and number of RBC units transfused if applicable. For biochemical evidence of hemolysis we considered haptoglobin<0.06, LD>1.5X ULN, presence of free hemoglobin in urine or plasma, indirect bilirubin>1.5x ULN. Results: During the study period, 891 adult doses of platelets were transfused. There were 43 (4.8%) out-of-group transfusions: 32 (3.6%) plasma-incompatible, 18 (2%) O platelets to non-O recipients (11 patients). DAT was performed in 5/11 patients. 2/5 had positive DAT. Both patients were blood group A and anti-A was eluted from their RBC. Both patients received 2 adult doses of platelets and experienced >10g/L drop in Hb however the biochemical evidence for hemolysis was inconclusive in both. One patient required a transfusion of 2u RBC. Conclusions: plasma-incompatible transfusion of platelets is rare. Group A patients and those receiving more than 1 dose of platelets appear to be at higher risk of having a positive DAT. Utility of routine DAT testing in this setting is questionable.

Keyword(s):
- platelet transfusion
- hemolysis
- DAT

Notes
Immunologic Abnormalities in a Triple Transgenic Mouse Model of Alzheimer Disease

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1Faculté de Pharmacie, Université Laval, 2Centre de recherche du Centre Hospitalier de l’Univ. Laval (CHUL), 3Département de R&D, Héma-Québec, Québec, Canada

IVIg was recently shown to benefit patients with Alzheimer disease (AD) in a phase 2 clinical trial, although the mechanism for this effect is currently unknown. Immunologic abnormalities have been described in patients with AD, in addition to the Aβ and Tau neuropathologies. The availability of an animal model that reproduces the neurological and immunological defects of AD pathology would therefore be an invaluable tool to study the mechanisms by which IVIg can ameliorate the condition of AD patients. In the present study, we evaluated whether the triple transgenic mouse (3xTg-AD) which reproduces the neuropathological hallmarks of AD, also reproduces the immunologic abnormalities observed in AD patients, using multiplex ELISA and cytometry analyses. A decrease in peripheral B (-42%, p=0.013) and T lymphocyte (-53%, p<0.001) numbers and an increase in CD4+/CD8+ T lymphocyte ratio (ratio from 1.47 for non-transgenic mice (NonTg) to 1.71 for the 3xTg-AD, p=0.018) associated with increased activation were observed in 12 month-old 3xTg-AD mice. Furthermore, we found that older 3xTg-AD mice exhibited a 5.5X decrease in IL-1α; (p<0.05) and a 5.7X increase in GM-CSF (p<0.05) plasmatic concentrations and higher IL-3 (2X, p<0.001), MIP-1α (2.6X, p<0.05) and IFNγ (2.2X, p<0.02) concentrations in the cortex compared to NonTg mice. Since most of these immunologic abnormalities are also observed in AD patients, we conclude that the 3xTg-AD mouse represents a suitable model to study the effects of IVIg in AD.
S. Ramirez-Arcos\*, S. Uzicanin\textsuperscript{1}, G. Clarke\textsuperscript{1}, and M. Goldman\textsuperscript{1}
\textsuperscript{1}Canadian Blood Services

Background: Since August 2010, blood donors at Canadian Blood Services are screened for hemoglobin (Hb) levels using the DiaSpect analyzer, a reagent-free full-spectrum photometer, in replacement of the qualitative copper sulphate method.

Purpose: To develop blood control samples to be used for periodic quality control (QC) verification of the DiaSpect Hb analyzer.

Methods: Commercial Hb controls, CPD-whole blood (WB) units, and EDTA venipuncture WB samples from 6 donors were evaluated in this study. Hb levels were measured daily, in triplicate, for 6 days, in 6 DiaSpect and one Advia®120 analyzers.

Results: Commercial Hb controls as well as CPD-WB units yielded lower and higher DiaSpect readings, respectively, when compared to Advia®120 measurements. EDTA WB samples were stable for the whole testing period and gave accurate results.

Conclusions: EDTA venipuncture WB samples stored for up to 6 days can be used in a QC verification program of the DiaSpect hemoglobinometer.

Keyword(s):
- DiaSpect
- hemoglobin
- hemoglobin analyzer
- donor hemoglobin
**Background and Purpose:** *Serratia marcescens* is a Gram negative bacterial pathogen that has been implicated in adverse transfusion reactions (ATRs) associated with contaminated platelet concentrates (PCs). The aim of this study was to investigate whether the ability of *S. marcescens* to form surface-attached aggregates (biofilms) might account for an increased ability to escape culture-based detection.

**Methods:** Five strains of *S. marcescens*, including biofilm-positive (BP) and biofilm-negative (BN) controls and three isolates associated with ATRs, were examined with respect to their capacity to grow in PCs and form biofilms. Clinical isolate 07-05 was selected to evaluate the link between biofilm formation and missed detection by the BacT/ALERT®3D system.

**Results:** All *S. marcescens* strains were able to grow in PCs. While only the BP control formed biofilms in conventional laboratory medium, four strains, including the BP and the three ATR-associated isolates, formed biofilms in PCs. Biofilms were comprised largely of dense aggregates of bacteria. The number of culture bottles detected for the clinical isolate 07-05 was ~2.5 times lower than the number of bottles detected for the BN control. Also, the average time of detection for biofilm-forming strain 07-05 was significantly delayed as compared to the BN strain.

**Conclusion:** The platelet storage environment promotes biofilm formation by *S. marcescens* likely increasing the risk of missed detection of this organism by conventional culture methods.

**Keyword(s):**
- *Serratia marcescens*
- biofilms
- platelet storage
- bacterial detection
C3  Prolonged Exposure to Room Temperature and it’s Correlation with Bacterial Growth in RBC Units

N. McLaughlin*, C. Sharpe, I. Sadek, D. Anderson, E. Kahwash
Canadian Blood Services, Halifax Centre, NS Canada

**Background:** Due to concerns of bacterial growth above 10°C, industry standards for four decades have allowed leukoreduced red blood cell units (LRBC) to be exposed to room temperature (RT) for a maximum of 30 minutes (min). Bacterial growth rate was measured in LRBC after prolonged exposure to RT.

**Method:** A total of 20 in-date LRBC were initially exposed to RT for 60 min then cooled (2-6°C). Group 1 (10) was exposed to RT for a total of 300 min over 3 periods; Group 2 was at RT for a total of 180 min over 2 periods. LRBC were cooled between each exposure. Core temperature (CT) was calculated from the surface temperatures of all sides of the unit bag at 120 min for all LRBC. Group 1 had aerobic bacterial testing (ABT) following two separate 120 min RT exposures; Group 2 after a single 120 min exposure.

**Results:** All 50 ABT samples were negative after 5 days incubation.

**Conclusion:** The results indicate that bacterial growth is unlikely in LRBC after prolonged exposure to temperatures which deviate from current standards, and that revision of the 30 min RT exposure rule may be considered.

**Keyword(s):**
- bacterial growth
- red blood cell
- room temperature
**Background:** Historically, red blood cells (RBC) are not to be outside of refrigeration for > 30 minutes (min) and must not exceed 10°C. This study will measure RBC warming rates and temperature (temp) variations.

**Method:** The 80 RBC units were removed from 2-6°C, bound in pairs, and a thermometer probe was placed in between (interior temp: int temp). The calculated core temp (CCT) was the average of the int and exterior (ext) surfaces. Temp was recorded at 0, 30, 60 and 120 min. Mean room temp was 20.6°C.

**Results:** The mean CCT upon removal from the fridge was 4.7°C. At 30 min it was 9.8°C (0.17°C/min warming rate). The mean CCT at 60 and 120 min was 12.3°C and 14.7°C, with a rate of increase by 0.083°C/min and 0.04°C/min, respectively. The difference between the two surfaces at 30 min was 5.1°C decreasing to 4.5°C and 2.0°C at 60 and 120 min, respectively.

**Conclusion:** The fastest rate of warming occurred in the first 30 min. Depending on the surface, method and technique used, the RBC temp could vary between 7.3°C and 12.4°C at 30 min in room temp, yet the CCT would be 9.8°C.

**Keyword(s):**
- rate of warming
- RBC temperature
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The Ottawa Hospital, Ottawa Hospital Research Institute and University of Ottawa, Ottawa, ON Canada

Background: Red blood cell (RBC) transfusion may prolong recovery in some patients, perhaps due to changes that occur during more prolonged RBC storage. We examined the impact of RBC transfusion and the age of transfused RBC units on clinical outcomes in hematopoietic stem cell transplantation (HSCT).

Study Design and Methods: Data concerning RBC transfusions between day 0 and day +30 were analysed for patients undergoing HSCT (n=555) at a single institution. “Old” RBC units were defined as those stored for 15 days or longer.

Results: Autologous transplant recipients (n=355, 3.8 units/patient) were more likely to avoid RBC transfusion and received fewer units compared with allogeneic recipients (n=200, 6.4 units/patient, p<0.0001). The mean number of transfused RBC units was greater in patients admitted to the intensive care unit (ICU) (10.5 vs 3.7 units/patient, p<0.01), correlated with longer length of stay (LOS) (p<0.0001) and correlated with increasing number of organ systems with toxicity ≥ grade 2 (p<0.0001). The proportion of “old” RBC units transfused and the mean age of transfused units did not correlate with 100 day non-relapse mortality (NRM), organ-specific toxicity, LOS, or incidence of ICU admission (p>0.05). In comparing the 71 patients who received only “old” RBC units with 218 patients who received only “new” RBC units, there was no increase in adverse clinical outcomes after HSCT.

Conclusion: Patients with increased RBC transfusion requirements have greater toxicity after HSCT. Whether RBC transfusion contributes to toxicity, however, remains unclear. The importance of RBC storage time appears negligible in HSCT.

Keyword(s):
- hematopoietic transplantation
- red cell transfusion
- age of blood
- clinical outcomes
HLA Antibody Prevalence in TRALI Associated Donors (TDS) Reported to CBS Since 2006

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**Purpose:** To compare HLA antibody prevalence in surveyed donors (SDs) and donors associated with cases likely to be TRALI (TDS).

**Methods:** TDS were donors associated with cases categorized as definite, possible and indeterminate by the TRALI Medical Review Group. Definite and possible TRALI cases were defined using the Canadian Consensus Conference definitions. Surveyed donors were deferred CBS donors from NEON and NetCAD that were enrolled in an antibody prevalence survey in 2007. Specimens were analysed for HLA class I and II antibodies at NBG ratios of 2.6 and 2.3, respectively.

**Results:** One hundred seventy-seven TDS and 149 SDs were studied. Higher prevalence of HLA antibody is observed in TDS compared to SDs (44.5% vs 22.1%, p χ²<0.001), with an identical trend for Class I (28.3% vs 16.1%, p χ²~0.01) and Class II (20.2% vs 6.0%, p χ²<0.001) respectively. Antibody specificity studies revealed a similar profile of HLA antigen in both populations.

**Conclusions:** TDS have higher prevalence of HLA antibodies than the SDs. However, many TDS did not have antibodies, suggesting that some cases, particularly in the indeterminate category, are not actually TRALI. Some cases may be due to anti-neutrophil antibodies and nonimmune mechanisms of TRALI. The frequency of HLA antibodies in SDs was similar to the REDS LAPS data. The high frequency of antibodies in SDs demonstrates the difficulty in basing a diagnosis of TRALI on lab data alone.

**Keyword(s):**
- HLA antibody prevalence
- survey blood donor
- TRALI associated blood donor

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Notes
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1University of Ottawa, 2St. Michael’s Hospital, 3University of Toronto.

Background: Cardiopulmonary bypass (CPB) during cardiac surgery is associated with platelet and coagulation defects including hypofibrinogenemia. In Canada, cardiac surgery is the most common indication for cryoprecipitate (CRYO) transfusion and yet there are no clinical studies to support its use in this setting.

Methods: We have evaluated respectively the prevalence of hypofibrinogenemia and CRYO transfusion post cardiac surgery on CPB at St. Michael’s Hospital. All adult patients undergoing CPB from January 1, 2010 to March 31, 2010 and who consented to blood product transfusion were enrolled in the study.

Results: 301 patient charts were reviewed and 293 patients met the eligibility criteria. Baseline characteristics were as follows: females 30% of patients, median age 67 yrs, median TRUST score 2, isolated CABG 58.7%, non-elective OR 19.4% 147/293 (50.2%) patients received allogeneic transfusion. 11/293 (3.8%) patients received CRYO within 24 hours of CPB, 5 of these were transfused CRYO intra-operatively. The patients who received CRYO were 82% male, median age 63 yrs, median TRUST score 2, isolated CABG 18% non-elective OR 45%. In 6/11 patients who received CRYO, pre-transfusion fibrinogen was <1.5g/L. 11/11 patients who received CRYO also received RBC and 10/11 also received FP. The reason for CRYO transfusion was “coagulopathy and bleeding” in all cases. Median dose of CRYO was 10 units.

Conclusion: Post-CPB CRYO transfusion rate of 3.8% is within the range of other Canadian institutions. Further analyses to investigate the association between hypofibrinogenemia following CPB and bleeding, blood transfusion, and adverse clinical outcomes are in progress.

Keyword(s):
- cardiopulmonary bypass
- cryoprecipitate
- fibrinogen
- transfusion

Notes

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Primary immunodeficiency diseases (PIDs) predispose patients to chronic and recurrent severe infections. Therapy for this condition requires lifetime immunoglobulin (IG) replacement. Subcutaneous IG (SCIG) is an alternative to IVIG that can be self-administered at home.

Purpose: To describe the real-life experience of patients with PIDs receiving SCIG therapy.

Methods: 5 adults and 1 child with PIDs who received home therapy with SCIG (Vivaglobin, CSL Behring) were described in the context of its efficacy in maintaining IG target levels, its tolerability, and its effects on quality of life.

Results: SCIG therapy had a positive impact on the patients’ daily life activities, was associated with a favourable side-effect profile, and was effective at maintaining IG target levels. Patients reported enjoying the flexibility of self-administration and an increase in the ability to travel.

Conclusions: SCIG therapy is especially ideal for those patients with difficult vascular access, intolerable severe side-effects with IVIG, and a busy and demanding lifestyle. In paediatric patients, SCIG therapy ameliorates the fear and anxiety of attending a medical facility.

Keyword(s):
- immunoglobulin
- immunodeficiencies
- subcutaneous
- self-administration
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**Purpose:** To investigate the cause of acute severe hemolysis in patients who had received Intravenous Immune Globulin (IVIG).

**Methods:** We have observed unexpected reaction in reverse grouping with A1 Cells and a positive DAT (Positive with anti IgG) in Group A and AB patients. Transfusion history was checked to resolve the discrepancy; it was found that patients have received IVIG. Acid Elution was performed. The Eluate was tested against A1 Cells, A2 Cells, O Cells (Screening Cells) and Auto Control.

**Results:** In a period of 1 year (between Dec 2009 – Dec 2010) we have seen about 10-15 cases of hemolysis due to IVIG. The following results were obtained:

<table>
<thead>
<tr>
<th>Type of IVIG</th>
<th># of patients</th>
<th>Blood Group</th>
<th>Diagnosis</th>
<th>DAT</th>
<th>Acid* Eluate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Privigen</td>
<td>6</td>
<td>5- A 1-AB</td>
<td>3 Kawasaki</td>
<td>Wk-3+</td>
<td>Wk – 3+</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2 ITP</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1 Sepsis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gamunex/ IGIvnex</td>
<td>3</td>
<td>2- A 1-AB</td>
<td>2 Kawasaki</td>
<td>Wk-1+</td>
<td>Wk-1+</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1 Sepsis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gammagard Liquid</td>
<td>2</td>
<td>2- A</td>
<td>2 Kawasaki</td>
<td>Wk-1+</td>
<td>Wk-1+</td>
</tr>
</tbody>
</table>

*Reacted stronger with A1 Cells than A2 Cells, negative with O Cells.

**Conclusion:** Passive anti-A from IVIG can cause positive DAT and acute Hemolysis in Group A and AB patients.

**Actions:** Issue group O red cells unless DAT and Eluate have been done and Anti A is not detected, to patients who are group A or AB and had received IVIG.

**Keyword(s):**
- IVIG transfusion
- acute hemolysis
- acid eluate
- passive Anti A
**Prothrombin Complex Concentrate Implementation and Use at a Community Hospital**

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Renfrew Victoria Hospital, Renfrew Ontario and The Ottawa Hospital, Ottawa, Ontario

**Background:** Prothrombin complex concentrate (PCC, Octaplex®) has been available from the Canadian Blood Services since fall 2008.

**Purpose/Methods:** (1) To describe the step-by-step approach to the successful implementation of PCC at a community hospital within a regional laboratory system. (2) To show the use of PCC after introduction at a community hospital. (3) To show the use of frozen plasma (FP) before and after PCC availability.

**Results:** (1) Before implementing PCC at the community hospital, an in-service was given to the hospital nursing, medical and laboratory staff, a written policy based on the NACBBP guideline was introduced and one laboratory technologist visited the central regional transfusion medicine (TM) laboratory to receive training in the reconstitution of PCC and in turn trained the rest of the laboratory technologists. (2) In the first year of PCC use (Jan – Dec 2009), 10 patients received PCC for emergency reversal of warfarin. In the second year (Jan – Dec 2010), 10 patients received PCC. Patient pre- and post-INR results and PCC dose will be presented at the meeting. (3) FP use at this community hospital is declining, with 20 units of FP issued in 2008 (pre-PCC), 18 units FP issued in 2009 and 17 units FP issued in 2010.

**Conclusions:** With a step-by-step approach, PCC can be successfully implemented in a community hospital within a regional laboratory system. Most PCC can be administered by the family physicians in emergency, with backup consultation with TM specialists for a few exceptional cases. Since implementation of PCC, FP use has decreased and the blood bank inventory of FP (group AB) has been reduced from 6 units to 4 units.

**Keyword(s):**
- prothrombin complex concentrate (Octaplex®)
- community hospital
- INR
- frozen plasma
Factors Affecting Platelet Transfusion in Cardiac Surgery

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1St. Michael’s Hospital, 2Li Ka Shing Knowledge Institute, 3University of Toronto, 4Canadian Blood Services, 5Sunnybrook Health Sciences Centre, 6University Health Network, 7Ottawa Health Research Institute, University of Ottawa

The objective for this study was to characterize factors associated with platelet transfusion in patients undergoing cardiac surgery.

Methods: This was a prospective study of all patients who had cardiac surgery over a 3-month period at St. Michael’s and Sunnybrook hospitals.

Results: 282 patients were included.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Patients transfused</th>
<th>Patients not transfused</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age (years)</td>
<td>N=51</td>
<td>N=231</td>
</tr>
<tr>
<td>Elective surgery</td>
<td>78.3%</td>
<td>83.4%</td>
</tr>
<tr>
<td>Emergent/urgent surgery</td>
<td>21.6%</td>
<td>16.4%</td>
</tr>
<tr>
<td>Mean preoperative platelet count (x 10(9)/L)</td>
<td>188</td>
<td>184</td>
</tr>
<tr>
<td>Preoperative use of ASA/thiopyridines</td>
<td>68.6%</td>
<td>88.8%</td>
</tr>
<tr>
<td>Mean intraoperative blood loss (ml)</td>
<td>1082</td>
<td>1336</td>
</tr>
<tr>
<td>Mean chest tube drainage (ml)</td>
<td>1229</td>
<td>1209</td>
</tr>
</tbody>
</table>

The mean intraoperative and postoperative platelet count at transfusion was 96 and 77 x 10(9)/L, respectively.

Conclusion: Future studies are needed to determine the optimal platelet count for transfusion.

Keyword(s):
- platelet transfusion
- cardiac surgery
The Rationale for Platelet Transfusion in Cardiac Surgery

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1St. Michael’s Hospital, 2Li Ka Shing Knowledge Institute, 3University of Toronto, 4Canadian Blood Services, 5Sunnybrook Health Sciences Centre, 6University Health Network, 7Ottawa Health Research Institute, University of Ottawa

Background: The rationale for platelet transfusion during cardiac surgery needs to be better defined as there is some evidence to suggest that platelet transfusion during cardiac surgery may be associated with adverse outcomes.

Methods: We developed a brief, valid and semi-structured questionnaire. We used focused physician interviews to determine the rationale for platelet transfusion in the perioperative setting. All anaesthesiologist and cardiac surgeons St. Michael’s Hospital and Sunnybrook Health Sciences centre were invited to participate.

Results: 23 physicians (18 anaesthesiologist, 5 cardiac surgeons) were interviewed over a 3-month interval. 40% of platelets were administered intraoperatively and 60% postoperatively. The rate and volume of bleeding perioperatively was reported as the most important factor for platelet transfusion followed by the platelet count and the type of surgery (i.e., emergent surgery). Preoperative use of anti-platelet agents, hemodynamic instability and duration of cardiac bypass were also selected as important factors for transfusing platelets.

Conclusion: There were consistent factors that influence physician’s decisions to transfuse platelets. Future studies need to define the need for transfusion for these indications and guidelines should better account for these factors.

Keyword(s):
- platelet transfusion
- cardiac surgery
- questionnaire

Notes
The Blood Utilization Program (BUP) at Vancouver General Hospital attempts to optimize preoperative hemoglobin in patients presenting for elective surgery. Referral criteria to our program include all patients presenting for major spine surgery, revision hip and knee replacement or cardiac surgery, patients with a preoperative hemoglobin ≤ 125g/L and patients refusing transfusion.

The purpose of this study was to measure the incidence of anemia in patients referred to the program and to measure the impact of interventions, including oral Fe, IV Fe and erythropoietin. All patients referred to the BUP and having surgery between May 1, 2010 and October 31, 2010 were included. BUP charts and lab and transfusion databases were interrogated to determine hemoglobin levels on referral to the program, treatment modalities used, and preoperative hemoglobin levels.

244 patients were referred during this period. The incidence of anemia (defined as Hb ≤ 125g/L) on referral to our program was 51%. The incidence of anemia preoperatively was 21%. The increment in hemoglobin level by intervention was also measured. In Fe deficient patients referred >2 months prior to surgery and receiving only oral Fe the average Hb increase was 22.5g/L. The average increase in Fe deficient patients receiving IV Fe was 17.7g/L. The average increase in patients receiving erythropoietin (many not Fe deficient) with oral +/- IV Fe was 13.3g/L.

In conclusion, our program successfully reduced the incidence of preoperative anemia. Fe treatment (oral and/or IV) used in Fe deficient patients resulted in the largest increase in hemoglobin level.

Keyword(s):
• anemia
• preoperative
• iron
• erythropoietin
Frequency of Thrombotic Events After Preoperative Erythropoietin in High Blood Loss Surgery: A Single Institution Audit

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Purpose: To determine the frequency of thrombotic events (TE) associated with the use of preoperative erythropoietin (EPO) for anemia optimization prior to high blood loss surgery at our institution.

Methods: Retrospective audit of all patients (pts) who received EPO in the Blood Conservation Clinic from January 2009 to December 15, 2010. EPO was administered at a dose of 40,000 units subcutaneously weekly to a target hemoglobin (Hb) of 120-130 g/L. Charts were reviewed to assess for any new documented TE (venous thrombosis, myocardial infarction, stroke) in the discharge summaries, laboratory values and radiology imaging in the 6 weeks after the procedure.

Results: During the audit period, 82 pts received 181 doses of EPO. The mean age was 66 yrs and 54 (66%) were women. The case mix was: 23 orthopedic, 22 cardiovascular, 22 gastrointestinal oncology and 15 gynecological oncology. The mean initial Hb was 107 g/L and a mean of 2.4 doses per pt was given. The mean preop Hb was 116 g/L (range 76-143 g/L). 9 pts had a preop Hb > 130 g/L. All 82 pts received thromboprophylaxis post-operatively. 3 pts had a venous TE (1 pulmonary embolus, 1 deep venous thrombosis and 1 with both) and 1 possible ischemic stroke. The preop Hbs for these cases were 105, 99, 128 and 130 g/L respectively. There was 1 death not felt to be related to EPO.

Conclusion: At our institution, EPO use for preop anemia optimization did not appear to result in an excess of thrombotic events when used to target hemoglobin of 120-130 g/L.

Keyword(s):
- blood conservation
- erythropoietin
- thrombosis
- anemia
Recombinant Erythropoietin and Risk of Allogeneic Red Blood Cell Transfusion in Neurosurgery

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Background: Pre-operative recombinant erythropoietin (EPO) has been shown to reduce the need for allogeneic red blood cells (RBC) and hasten erythroid recovery post-operatively in a variety of surgical setting. However, data on the efficacy of EPO in neurosurgery is lacking.

Methods: We performed a case control study examining whether preoperative EPO in neurosurgical patients reduces allogeneic RBC transfusion. Between 2002-2009, fifty six neurosurgical patients (43 females, 50.50±13.56 years) received re-operative EPO at St. Michael’s Hospital through the ONTraC provincial blood conservation program. Patients received 1-4 EPO injections (means 2.38±1.01, 40,000 U/dose) prior to surgery. Cases were matched 2:1 for age, sex and ICD code most responsible for surgery (N=112 control patients, 86 females, 51.09±13.99 years). The primary outcome was allogeneic RBC transfusion.

Results: Thirty patients receiving EPO (53.6%), but no control patients, donated autologous blood prior to surgery (PAD). Of these, 16 patients (53.3%) received autologous blood. Peri-operatively, 10 index cases (17.9%) and 20 control cases (17.9%) received allogeneic RBC. The number of allogeneic RBC transfused did not differ between groups (0.63±1.66 erythropoietin cases vs. 0.44±1.38 control patients, Z=0.14, p=0.884)). When both autologous and allogeneic transfusions were considered, patients receiving EPO were significantly more likely to have received RBC (1.07±1.94 EPO cases vs 0.43±1.38 control patients, Z=2.51, p=0.012). Of those patients who received EPO and allogeneic RBC, 70% had predonated autologous blood (1.57±78 units). Limitations: This is a retrospective study that considered only EPO and PAD in blood conservation.

Conclusion: EPO did not reduce the risk of allogeneic RBC transfusion in neurosurgical patients. When combined with PAD, an increased risk of RBC transfusion was noted in this population.

Keyword(s):
• blood conservation
• recombinant erythropoietin
• neurosurgery

Notes
Can OrthoPAT\textsuperscript{R} Reduce Allogeneic Transfusion At Arthroplasty?

Adrienne Carr MD, Rebecca Rock RN\textsuperscript{*,} Charles MacAdams MD FRCPC

**Purpose:** Purpose was to estimate whether OrthoPAT\textsuperscript{R}, a centrifugation-based intra- and postoperative red blood cell salvage system, could reduce allogeneic transfusion at arthroplasty.

**Methods:** The Health Research Ethics Board provided approval for patient chart review if required. We prospectively recorded volumes of red blood cells salvaged with OrthoPAT\textsuperscript{R} and rates of allogeneic transfusion in 203 consecutive arthroplasty patients of one surgeon from 2008 to 2010. We estimated what allogeneic transfusion rates may have been for 2 hemoglobin transfusion thresholds had OrthoPAT\textsuperscript{R} been unavailable. For calculations, we used an OrthoPAT\textsuperscript{R} product hematocrit of 75 and estimated patient blood volume of 5 L.

**Results:** For all patients, mean preoperative and nadir hemoglobin levels were 139 g/L and 104 g/L. Eleven of 203 patients (5.4\%) received allogeneic transfusion. Mean OrthoPAT\textsuperscript{R} volumes returned to patients were 371 ml (knee) and 226 ml (hip). Estimated nadir hemoglobins had OrthoPAT\textsuperscript{R} been unavailable were 87 g/L (knee) and 93 g/L (hip). Estimated mean transfusion rates for all patients had OrthoPAT\textsuperscript{R} been unavailable were 14.8\% at transfusion threshold 70 g/L and 32.5\% at transfusion threshold 80 g/L.

**Conclusions:** Estimated mean nadir hemoglobin levels without use of OrthoPAT\textsuperscript{R} (87 – 93 g/L) were only slightly lower than those actually achieved (104 g/L). But for numerous patients, OrthoPAT\textsuperscript{R} was vital to avoidance of transfusion thresholds, and was estimated to have decreased transfusion rates by 64 – 83\% depending on transfusion threshold of 70 versus 80 g/L.

**Keyword(s):**
- blood transfusion
- arthroplasty
- blood salvage
- perioperative anemia

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**Notes**
Anti-D Immunization by RH DEL Type RBCs

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Background: An 88-year old D- woman with no previous transfusions and 1 pregnancy had a negative antibody screen. 4 weeks after receiving 2 units of O, D- RBCs, I.S. compatible, her antibody screen tested positive and anti-D was identified. Both transfused units phenotyped as D-, C+, E-, c+, e+. An investigation was initiated.

Methods: Extended serologic D antigen testing included antiglobulin testing for weak D; the ALBAclone® Advanced Partial RhD Typing Kit; and adsorption-elution with Immucor polyclonal anti-D reagent. RHD genotyping was performed by Progenika Inc.

Results: 1 donor confirmed as D-. 1 donor tested negative by the weak D test and ALBAclone® Kit but was determined serologically to be DEL by adsorption-elution and genotyped as RHD94InsT (FS, 35X), an RHD allele known to encode a DEL phenotype. Lookback showed that of 3 other RBC units transfused, 2 recipients remain antibody negative and 1 was lost to follow-up.

Conclusion: Transfusion of D DEL RBCs, missed in routine testing, resulted in anti-D immunization of 1 of 3 recipients tested. The polyclonal anti-D used in this case is an appropriate reagent to detect extremely weak D variants by adsorption-elution. Molecular testing confirmed rare serological findings.

Keyword(s):
- immunization
- RH DEL
- adsorption-elution
- genotype
Background: The LW and LWb antigens are part of the Landsteiner Weiner blood group system. Anti-LW and anti-LWb antibodies are usually IgG but can be IgM, allo or auto in nature and may cause HDN and/or transfusion reactions. The LW and Rh genes are independent, but the LW glycoprotein is part of the Rh membrane complex. D+ RBCs give stronger reactions than D- RBCs with anti-LW. Anti-D may be misidentified due to weak reacting anti-LW. Case Report: 1 case of alloanti-LWb confirmed and 2 cases of questionable anti-D in D+ individuals were examined. All plasmas were strongly reactive with D+ and non-reactive with D- RBCs by solid phase methods and confirmed by NIRL using standard tube IgG methods. Additional testing performed by NIRL showed enhanced reactivity by PIAT and no reactivity with 2M DTT treated RBCs by IAT. Rare LW(a-b-) RBCs tested by IAT were non-reactive. When possible, LWa and LWb phenotyping was performed. In the case of the propositus, the patient phenotyped as LW(a-b-) and apart from a sibling is the only other known genomic LW(a-b-). In the 2 cases of auto-LWa, the patients phenotyped as LW(a+) 3+ and LW(a+) weak.

Conclusion: Treatment of RBCs with papain, trypsin, chymotrypsin, pronase, neuraminidase and 2 M DTT or AET in addition to plasma adsorptions, rare RBCs and antisera will aid in the distinction between anti-D and anti-LW.

Keyword(s):
- alloanti-LWb
- anti-D
- auto-LWa
- 2M DTT/AET
Chimera – Blood Group Discrepancy

S. Jivraj*, Z. Salomon de Friedberg, W. Lau
CBS Central Ontario – Diagnostic Services

Purpose: To demonstrate the importance of patient history and good communication between Transfusion Medicine departments in resolving blood group discrepancies. A sample from a 30 year old patient with diagnosis of von Willebrand’s disease and colon cancer was referred to our Reference Laboratory for investigation of ABO discrepancy and/or possible weak subgroup of A.

Method: Patient’s plasma was tested with anti-A, anti-B and anti-A,B at immediate spin and then incubated at R.T. for thirty minutes. Patient’s red cells were papainized and tested with anti-A, anti-B and anti-A,B.

Results: The ABO grouping results by our laboratory were as follows:

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>-A</th>
<th>- B</th>
<th>-A, B</th>
<th>- A1 cells</th>
<th>B cells</th>
</tr>
</thead>
<tbody>
<tr>
<td>Immediate spin</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>After 30 min RT incubation</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>After papain treatment of all cells</td>
<td>3</td>
<td>0</td>
<td>3</td>
<td>0</td>
<td>4</td>
</tr>
</tbody>
</table>

Adsorption/elution studies were performed and anti-A could not be eluted from the patient’s red cells. These serological results are not consistent with any known subgroup of A.

Conclusion: Further information was requested from the referring hospital and we learned that the patient had received a stem cell transplant at age 13. Records from the pediatric hospital indicated that the patient was originally group A and received an allogeneic bone marrow transplant in 1994 from a group O donor. The patient is most likely a chimera with the patient’s original progenitor cells continuing to produce small amounts of group A cells.

Keyword(s):
- chimera
- patient’s history
- blood group discrepancies
- bone marrow transplant

Notes
Background: A 70 y old Afro-Caribbean female suffered rapid multi-alloimmunization on initiation of transfusion support for myelofibrosis. Several antibodies were identified as being specific for antigens which she herself was phenotype-positive for. To resolve discrepancies, genotyping was undertaken.

Studies and Method: Serology was performed by standard testing. RHD and RHCE genotyping was performed by PCR-multiplex, PCR-RFLP, and AS-PCR analysis for specific exons, with zygosity determination by hybrid box detection.

Results: The patient originally typed as blood group O with mixed-field Rh(D) expression, while anti-E was found on screen. A week after receiving 2u of E- RBC, a delayed serologic transfusion reaction occurred due to anti-Fy(a) and anti-D. Another week later, anti-C emerged, and 3 wks after this time, anti-Jk(b) was detected. The original patient phenotype was D+C+E-c+e+Fy(a-)b-) Jk(a+ b-), which concurred with the anti-E, -Fy(a), and -Jk(b), but which disagreed with the anti-D & -C. To decipher the nature of the latter, RH genotyping showed that the RHD alleles consisted of a RHD-CE-D hybrid (encoding only altered C), and weak partial RHD type 4.0, while the RHCE alleles in turn were RHCE*ce\(^2\) homozygous, reflecting altered/partial e. We decided to transfuse D-C-E-K-Fy(a-) Jk(b-) units, despite potentially provoking anti-e or anti-V/hrB, rather than source r’S homozygous units. The transfusion trigger was lowered to <70g/L. She has since tolerated 39 such units without reactions or new antibody formation.

Conclusion: Genotyping helped to resolve a discrepancy and informed a risk-benefit decision on the safest and most accessible blood for transfusions.

Keyword(s):
- genotype
- red cell alloimmunization
- partial alleles
- Rhesus (Rh)
Case Report: Direct Agglutinating Antibody as Cause of ABO Discrepancy

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CBS Central Ontario – Diagnostic Services

Purpose: To demonstrate the importance of the Immediate Spin (IS) phase of the Saline Indirect Antiglobulin Test (SIAT) when investigating an ABO discrepancy in the presence of an alloantibody. Samples from a previously transfused 65 year old male being prepared for the OR were referred to our reference laboratory for investigation.

Method:
ABO testing:

<table>
<thead>
<tr>
<th></th>
<th>-A</th>
<th>-B</th>
<th>-A, B</th>
<th>A, cells</th>
<th>B cells</th>
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<tbody>
<tr>
<td>IS</td>
<td>0</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>1</td>
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</tbody>
</table>

The patient was confirmed group B. Antibody investigation identified Anti-Fya and Anti-S by MTS Gel. Investigation proceeded to determine if discrepant ABO results may have been caused by the identified alloantibody:

<table>
<thead>
<tr>
<th></th>
<th>S-, Fya-</th>
<th>S-, Fya+</th>
<th>S+, Fya-</th>
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</thead>
<tbody>
<tr>
<td>IS</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>30 min/RT</td>
<td>0</td>
<td>0</td>
<td>w</td>
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<tr>
<td>30 min/37°C</td>
<td>0</td>
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<tr>
<td>AHG</td>
<td>0</td>
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</table>

Reverse grouping discrepancy resolved by:

<table>
<thead>
<tr>
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<th>-B</th>
<th>-A, B</th>
<th>A, cells</th>
<th>B cells</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 min/37°C</td>
<td>0</td>
<td>4</td>
<td>4</td>
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<td>0</td>
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</table>

Conclusion: ABO discrepancy may be caused by a Direct Agglutinating Antibody (DAA). In this case, anti-S is acting as a DAA. Anti-S is known to demonstrate optimal reactivity at temperatures below 37°C. Tests performed below 37°C may help to identify antibody specificity for anti-S.

Keyword(s):
- direct agglutinating antibody
- ABO discrepancy
- immediate spin

Notes

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L. Harrison1*, J. Zeng1, B. Hannach2
1The Credit Valley Hospital, Mississauga ON Canada, 2Canadian Blood Service, Toronto Centre, Toronto ON Canada

Purpose: A patient admitted through the Emergency Department tested O Pos with mixed field Rh grouping, positive antibody screen, and positive direct antiglobulin test. The patient had been admitted 5 days prior for ruptured ectopic pregnancy and had received 2 O Negative and 2 O Positive units of uncrossmatched red cells.

Method: Antibody panels showed possible Anti-D specificity. This was confirmed by the eluate showing Anti-D specificity. The initial antibody screen had been negative. Rh grouping results of 4+ did not indicate a weakD or partial D phenotype, likely to produce an immune Anti-D. In June 2010, Canadian Blood Services (CBS) began issuing red cell products from donors with blood group antibodies, with antibodies documented on the label. Our facility decided to handle such donor units as routine stock with no documentation of antibodies present therefore no antibody information was available for the transfused donor units. A transfusion reaction report (delayed hemolytic) was completed and CBS was notified, including a request for donor unit information.

Results: CBS confirmed that the donor of one of the transfused O Negative units had Anti-D, Anti-C, and Anti-s.

Conclusion: The antibody was reported as Passive Anti-D due to antibody in the donor unit. Based on this case, do we need to alter our policy?

Keyword(s):
• uncrossmatched blood
• donor antibody
• transfusion reaction
• passive Anti-D

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Notes
**C23  Anti-D is the Most Common Alloantibody in Female Blood Donors in Nova Scotia**

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Canadian Blood Services Halifax NS

**Purpose:** To obtain the relative frequencies of red blood cell (RBC) antibodies in blood donors in Nova Scotia.

**Methods:** All blood donor files for the period Jan-2005 to Jul-2010 were reviewed; files with positive antibodies were recorded.

**Results:** Total donor files were 51,135; 54% male, 46% female. A total of 147 donors (0.29%) were found to have antibodies. Of them 73% were female. (M:F alloimmunization rate of 1:2.7). The five most common alloantibodies were: anti-D (18%), anti-K (16%), anti-E (14%), anti-M (11%), and anti-C (5%). Females represented 81% of the occurrence of anti-D. Anti-K was similarly divided between genders (79% female; 21% male), as was anti-E (95% female; 5% male). Anti-D and anti-C was the most common coexistent antibody pair (at 4% of alloimmunized donors), with two-thirds present in female donors.

**Conclusion:** The finding of anti-D being the most common antibody in females is of interest considering the widespread use of RhIg over the past 4 decades which has lead to decreased rates of sensitization to the D antigen based on other published studies.

**Keyword(s):**
- antibody
- blood donor
- Anti-D
Impact of CBS Antigen Typings on a Transfusion Service

H Blain*, K Hamacher, G. Clarke, S. Nahiriak. Alberta Health Services - Edmonton

**Background:** Our regional transfusion service locates antigen (Ag) negative RBC units for patients with antibodies by review of regional inventory or from units typed by Canadian Blood Services (CBS). Units received have historically been retyped for ABO and Rh, as well as any applicable RBC antigens before serological crossmatch for patients with antibodies. New initiatives in Ag typing by CBS have resulted in an increasing number of pre typed and labeled units in our inventory. We have re evaluated the necessity of confirming the Ag type of units end labeled by CBS for serological crossmatch without retesting for the non ABO Ags.

**Method:** Over a two month period, rbc units were retested for ABO and Rh with phenotypes of units end labeled by CBS entered into the laboratory information system (LIS), without confirmatory typings, at the time of inventory receipt. Data was gathered for all Ag negative units required to determine the number of units with known phenotypes provided by CBS, type and frequency of Ags tested and savings of time and reagent costs.

**Results:** Over a 2 month period 666 units needed Ag typing for 1271 antigens. Of these 1271 Ag types, 539 (42.4%) were done by CBS and were not repeated. The most common Ag typings required were Kell (K) (24.3%), Kidd (Jka) (16.8%) and Rh E (21.4%). CBS typed units were identified in our inventory 32.7%, 22.0% and 65.1% of the time respectively. The average time required for entry of labeled typings is 30 seconds per Ag versus 20 minutes for typing - saving approximately 4.5 hours in the 2 month period. The cost savings for the new process during the two months (reagent +technologist time) was $3734.40.

**Conclusions:** Provision of rbc units labeled with known phenotype is of significant benefit to the hospital transfusion service.

**Keyword(s):**
- antigen typing
- cost savings
Dana L. Kyluik*, Wendy M. Toyofuku and Mark D. Scott
Canadian Blood Services & University of British Columbia

Background: The immunocamouflage of allogeneic donor cells has primarily focused on the covalent grafting of methoxypoly(ethylene glycol) [mPEG] to cell membranes. The grafted mPEG dramatically reduces in vivo antigenic recognition and immunogenicity of the donor cells. However, a small body of literature may argue against clinical use of mPEG. Polyoxazoline [POZ] polymers may be an effective next generation alternative to mPEG.

Methods: To assess the utility of POZ, human red blood cells (RBC) and peripheral blood mononuclear cells (PBMC) were modified with activated 20 kDa POZ or mPEG (0-4mM; 60 minutes at pH 8.0). A two-phase separation system was used to quantify RBC membrane modification. RBC osmotic fragility was used to assess membrane stability. PBMC viability was assessed via 7-amino-actinomycin D (7-AAD) incorporation. The efficacy of immunocamouflage was assessed by reduced detection of blood group antigens and PBMC CD markers via flow cytometry.

Results: Grafting of mPEG and POZ to RBC resulted in comparable 2-phase partitioning curves. mPEG showed improved partitioning over POZ at lower grafting concentrations (0.5 mM 36.2±6.1 vs 11.2±2.8%), while POZ displayed slightly superior partitioning at higher concentrations (2 mM 59.9±3.5 vs 73.2±5.0%). Osmotic fragility revealed no overall differences in RBC membrane stability while POZ demonstrated slightly elevated spontaneous lysis at ≥ 2 mM. Both polymers showed similar dose dependent decreases in PBMC CD3 and CD4 detection. At 2 mM, CD3 and CD4 detection was reduced 88% and 98% with mPEG and 86% and 97% with POZ.

Conclusions: This data clearly demonstrates that other polymers can effectively modulate the antigenicity and immunogenicity of allogeneic donor cells. Successful implementation of this technology may prove useful in preventing rejection of allogeneic blood cells and tissues.

Keyword(s):
• red blood cells
• white blood cells
• immunocamouflage
• alloimmunization

Notes
Iron Overload Causes Immunological Dysfunction

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Canadian Blood Services and the Centre for Blood Research and the Department of Pathology and Laboratory Medicine at the University of British Columbia, Vancouver, BC, Canada

**Background:** Primary and secondary iron overload leads to increased morbidity and mortality due to redox-mediated tissue damage. However, the effects of excess iron overload on immune function are less understood.

**Methods:** To determine the effect of iron overload on adaptive immunity, antigen presentation assays were used. Dendritic cells (DC) were pre-incubated with iron and challenged with tetanus toxoid (TT) and then cocultured with autologous peripheral blood mononuclear cells (PBMC). PBMC were labelled with 5,6-carboxyl-fluorescein diacetate succinimidyl ester (CFSE) to measure cell proliferation via flow cytometry. The effects of iron chelators such as Desferal (DFO) and Deferiprone (L1) were assessed. To demonstrate the molecular mechanism of the iron overload in dendritic cells, mitochondrial potential was detected using JC-1 distribution assay.

**Results:** Iron overload significantly inhibited antigen presentation (TT and RhD) as evidenced by significantly decreased PBMC proliferation. Pre-treatment of DC cells with 200 µM FAC for 24 hours resulted in a ~65.2% reduction in PBMC proliferation in response to the TT following 14 days incubation. As expected, iron chelators (e.g., 200 µM DFO or L1) restored the PBMC proliferation in a concentration dependent manner. The iron overloaded DC demonstrated 26.3% mitochondrial membrane depolarization while inclusion of L1 and DFO resulted in mitochondrial membrane potentials similar to healthy control cells.

**Conclusions:** As demonstrated, iron overload results in a significant dysfunction of the immune system. Iron chelation effectively restored the immune response partially through the recovery of mitochondrial membrane potential in the dendritic cells. This data suggests that proper chelation of primary and secondary iron overload states is necessary for the maintenance of a patient’s immune competency.

**Keyword(s):**
- iron
- secondary iron overload
- antigen presentation
- iron chelators

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Notes

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INVEST IN YOUR FUTURE

CSTM 2011 - 41
Suppression of Protein Phosphatase 2A Activity Enhances Ad5/F35 Adenovirus Transduction Efficiency in Human Normal B Lymphocytes and Cell Lines

MP. Cayer1, M. Samson1,2, C. Bertrand1,2, N. Dumont1, M. Drouin1, and D. Jung1,2 *
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Adenoviral vectors have the particularity of efficiently and rapidly transduce both proliferating and quiescent cells and do not integrate into the host genome, allowing a reproducible gene expression level. However, we previously demonstrated that Ad5/F35 vectors transduce plasma cell lines at higher frequency than primary B cells, mainly through difference in intracellular trafficking. Since phosphatases are well known to influence intracellular trafficking, we analyzed here the effects of different phosphatase inhibitors on transduction efficiency in B lymphocytes. Inhibition of PP1 by Tautomycin or PP2B by cyclosporine A had no effects on adenoviral transduction efficiency whereas inhibition of PP2A with Cantharidic acid or PP1 and PP2A with Okadaic acid substantially increase transduction efficiency. Moreover, inhibition of PP2A results in a rapid increase of AKT, ERK1/2 and MEK1/2 phosphorylation. Importantly, confocal microscopy analyses revealed that inhibition of PP2A abolishes the recycling of adenovirus and enhances their escape from early endosome to the nucleus. Finally, no increase of adenoviral entry was observed following PP2A inhibition. Our results are in accord with reports indicating that PP2A is involved in the formation of recycling vesicles and may improve gene transfer in the context of gene therapy.

Keyword(s):
• gene transfer
• adenovirus vectors
• B lymphocytes
• phosphatase inhibitor
Distinctive Effects of Various Antioxidants on Phenotypes of CD19+ and IgG+ B Cells

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Héma-Québec, Québec, Canada, G1V5C3

Purpose: Our laboratory focus on optimizing in vitro IgG production by cultured human B cells as a substitute for IVIg for which demand over the last seven years in Canada has increased by 64%. Here, we have analyzed the effect of REDOX homeostasis imbalance on B cell maturation and production of IgG.

Method: Purified CD19+ B cells were treated with various antioxidants: N-acetylcysteine (NAC), α-tocopherol (α-toco) and Trolox (Tlx) as purified IgG+ B cells were only treated with NAC. Cell counts and viability were determined with Trypan blue exclusion. Flow cytometry analysis was done on viable (Pacific Blue negative) and CD19+ or CD45+ cells. The relative concentration of Reactive oxygen species (ROS) was determined with the probe CM-H2DCF-DA (DCF) and intracellular signaling was determined with Fluorescent Cell Barcoding (FCB). IgG secretion was measured by ELISA. Pictures were taken with a Retiga 1300 camera (Q-Imaging) combined to a Nikon Eclipse TE2000 microscope.

Results: All antioxidants decreased intracellular ROS concentrations in B cells. NAC treatment decreased expansion and secretion of both CD19+ and IgG+ cells and increased viability and homotypic adhesion of CD19+ cells. Besides, Tlx and α-toco treatments had no effects on CD19+ B cell functions and phenotypes. Flow cytometry analysis of intracellular signaling showed that NAC, but not Tlx and α-toco, inhibits STAT3 phosphorylation in CD19+ cells. Finally, NAC also inhibits STAT3 in IgG+ cells.

Conclusion: Our results indicate that B cell REDOX homeostasis has specific role during B cell differentiation. Furthermore, our results suggest that NAC may also have antioxidant-independent functions such as direct targeting of specific signaling pathway.

Keyword(s):
- B lymphocyte
- REDOX
- IVIg
- signaling

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Notes

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RBC Membrane Fragility as an Age-Independent Metric of Stored Blood Quality

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\textsuperscript{1}University of Michigan, Ann Arbor, MI, USA; \textsuperscript{2}Blaze Medical Devices, Ann Arbor, MI, USA

Long storage times for blood products are often unavoidable. Product age is essentially the only indicator used today for Red Blood Cell (RBC) quality loss during storage. Much controversy persists over the impact of RBC age on transfusion outcomes, as studies on this issue remain inconclusive. Such inconsistency may arise from unit-to-unit variability (e.g. due to donor-to-donor differences), which likely introduces some age-independence to RBC storage lesion and thus post-transfusion performance. Hence, quality metrics other than storage time could aid with inventory management and/or treatment decisions. RBC membrane mechanical fragility is proposed as one such candidate in vitro metric: it aggregately reflects a range of biochemical and biomechanical changes associated with storage lesion, and can provide a more comprehensive characterization of particular units than other properties. Subjecting RBC to shear stresses of multiple varying parameters (such as stress magnitude and duration) can generate multidimensional fragility profiles able to capture more detailed information than with single-parameter or single-point measurements. Preliminary data suggest RBC fragility profiles can vary substantially among units of equal age, and current work is investigating the correlation between RBC in vitro fragility profiles and cell post-transfusion survival in vivo.

Keyword(s):
- RBC
- transfusion
- storage lesion
- membrane fragility


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Notes
Novel Severe Factor V Deficiency

K. Talbot\textsuperscript{1,2,3}, J. Song\textsuperscript{1,2,3}, J. Tamura-Wells\textsuperscript{1,2,3}, L. Eghdam\textsuperscript{1,2,3}, J. Hewitt\textsuperscript{2,4}, L. Vickars\textsuperscript{5}, C.J. Carter\textsuperscript{2,3}, R.T.A. MacGillivray\textsuperscript{2,4}, E.L. G. Pryzdial\textsuperscript{1,2,3}

\textsuperscript{1}CBS, R&D, \textsuperscript{2}UBC/Centre for Blood Research, \textsuperscript{3}Dept. Pathol.Lab. Med., UBC, \textsuperscript{4}Dept. Biochem. Mol. Biol., UBC, \textsuperscript{5}Hematology, St. Paul's Hosp., Providence Health Care

Congenital deficiency of plasma factor V (FV) is a rare clotting disorder that can result in a severe bleeding diathesis. Conventional treatment is with frozen plasma. In this study a FV deficient patient aged 73 exhibited severe bleeding tendency due to minor trauma. Other clotting factors were normal. Based on the lack of parental bleeding, compound heterozygosity in the FV gene was probable. The patient F5 gene (exons and intron/exon junctions) was sequenced. Both wild type (WT) and mutated FV genes were cloned into expression vectors, and recombinant (r) FV proteins were generated. Patient plasma and rFV were analyzed by enzyme immunoassay, clotting assays and western blot. DNA sequence analysis revealed two novel mutations; Leu1821 to Ser (L1821S) and Gly2192 to Cys (G2192C). Patient plasma FV activity was low; 0.5% (extrinsic) and 2% (intrinsic) with only 9% of the normal antigen level. Compared to WT, rFV activity and secretion were also low, L1821S (15/28%) and G2192C (0/25%). Thrombin pre-treatment cleaved all FV proteins and enhanced clotting activity for most. In conclusion, two novel FV mutations that affect both functions and secretions were revealed. L1821S is located close to a FV metal ion binding site, which may explain reduced function and secretions. G2192C introduces a thiol (known to affect protein folding) to FV, and may explain attenuated secretion and undetectable activity. These studies contribute to understanding genotype to phenotype correlations of FV deficiency with a goal to improve blood product utilization strategy and patient outcome.

Keyword(s):
- factor V
- plasma
- coagulation
- deficient

\textbf{Notes}

Purpose of investigation: We previously showed that the efficacy of antibiotic disinfection of allografts valves was improved when carried out at 37°C instead of 4°C. In this study, we have compared the impact of the antibiotic disinfection temperature on the structural integrity of heart valves.

Methods: Porcine heart valves (n=40) were processed according to our human heart valve allograft procedures. Valves were randomly distributed into 2 groups and treated with an antibiotic cocktail (vancomycin, gentamicin and cefoxitin) for 24 hours. One arm was treated at 4°C, the other at 37°C. Valves were frozen in DMSO and stored in liquid nitrogen. After thawing, one leaflet from each valve was incubated in a culture medium to evaluate cellular viability. The second leaflet was used to measure mitochondrial activity with the MTT assay, and the last leaflet was used for collagen determination.

Results: After thawing, cell viability and cellular counts were similar for both treatments (p>0.05). Collagen content and mitochondrial activity were slightly but statistically higher in valves treated at 4°C (349 ± 254 vs. 202 ± 206 µg/g for collagen (p=0.049) and 5.14 ± 1.07 vs. 4.01 ± 1.31 OD450nm/g for the MTT assay (p=0.005).

Conclusion: These findings suggest that a warmer temperature of disinfection has only minor effects on the structural integrity of porcine heart valves. The question remains as to whether the same is true for human allografts.

Keyword(s):
- heart valve
- tissue banking
- process development
- disinfection

Notes
Pathogenesis of Fetal and Neonatal Immune Thrombocytopenia: Contribution of Impaired Angiogenesis

Lang, S.1,3,4*, Yang, H.3,4, Boyd, S.4, Chen, P.3,4, Zhao, X.4, Li, C.1,4, Piran, S.1,4, Freedman, J.1,2 and Ni, H.1,2,3,4
1Depts. of Laboratory Medicine and Pathobiology, and 2Medicine, University of Toronto, 3Canadian Blood Services; 4St. Michael’s Hospital

Fetal and neonatal immune thrombocytopenia (FNIT) is a severe bleeding disorder which results from fetal platelet opsonization by maternal antibodies and subsequent platelet destruction. Two major platelet antigens targeted by antibodies in FNIT are GPIbIlla (αIIbβ3 integrin) and GPIbα. It has been reported that angiogenic endothelial cells (ECs) express β3 integrin on their surface and that β3 integrin is required for angiogenesis. Therefore, we investigated whether anti-β3 integrin (anti-β3) antibodies in FNIT cross-react with blood vessels of the developing fetus/neonate and contribute to pathogenesis. Antibodies to GPIbα (note expressed on ECs was used as a control.

Results: β3 integrin- or GPIbα-deficient female mice were bred with wild-type male mice to generate heterozygous fetuses. The fetuses induced maternal immune responses as observed in human FNIT. Both groups had reduced platelet counts but ICH was only observed in some fetuses/neonates with anti-β3 antibodies. Pups with anti-β3 antibodies had increased apoptosis in the brain. Blood vessel development was examined by immunostaining both the brain and retinal vasculature. Anti-β but not anti-GPIbα, antibodies impaired vascularization of both organs. Maternal IVIG treatment could ameliorate these complications. In addition, anti-β3 sera inhibited tube formation by ECs in vitro.

Conclusion: Anti-β3 integrin antibodies in FNIT likely impair angiogenesis in the developing fetus/neonate. This impairment can be ameliorated by maternal IVIG treatment.

Keyword(s):
• FNIT
• IVIG
• platelets
• angiogenesis
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Canadian Blood Services and St. Michael's.

There are many theories as to the mechanism of action of IVIg in the treatment of autoimmune diseases such as ITP. One prominent theory involves accelerated pathogenic autoantibody clearance by saturation of the neonatal Fc receptor (FcRn). FcRn is an Fcγ receptor involved in binding serum IgG, increasing its half-life by rescuing it from catabolism. In the treatment of ITP, it has been theorized that IVIg saturates FcRn, blocking the ability of anti-platelet antibodies to bind FcRn and increasing their catabolism, thus decreasing their ability to mediate platelet clearance. Both IVIg (2 g/kg body weight) and monoclonal antibodies to CD44 (2 mg/kg body weight) successfully ameliorate murine ITP. To definitively determine if the neonatal Fc receptor (FcRn) is required for the acute amelioration of ITP by these two therapeutics, we employed FcRn deficient mice in the murine ITP model. Here, we demonstrate that FcRn deficient mice treated with IVIg or a CD44 antibody (at a 3 log fold lower dosage than IVIg) were protected from ITP to the same extent as wild-type mice. FcRn has an absolute requirement for the protein β2 microglobulin (β2M) to be functionally expressed. To verify and substantiate the results found with FcRn deficient mice, we next employed β2M deficient mice in the murine ITP model and found these mice treated with IVIg or KM114 were also protected from ITP. These data suggest that for both high dose IVIg as well as low dose monoclonal CD44 antibody treatment in an acute ITP model, FcRn expression is dispensable.

Keyword(s):
• ITP
• monoclonal antibody
• IVIg
• FcRn

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Notes

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Mechanisms Contributing to Cytopenias Mediated by an Anti-RBC Antibody TER119

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1Canadian Blood Services, 2St. Michael’s Hospital, 3Department of Lab Medicine and Pathobiology, Univ. of Toronto

Introduction: The monoclonal antibody (Ab) TER119 recognizes a molecule associated with glycophorin A on the surface of mouse red blood cells (RBC). It was previously demonstrated by our group and others that TER119 has an anti-D-like effect in that it ameliorates passive immune thrombocytopenia (ITP) in a mouse model, although the mechanism of action remains unclear. We therefore initiated work in this study to investigate the pathologic and therapeutic properties of TER119.

Methods: The anemic effect of TER119 was studied 1) in vitro; and 2) in vivo, by injecting the Ab into mice knocked-out for selected genes of interest.

Results & Conclusions: TER119 injected alone induced significant anemia in 3 strains of standard inbred mice (C57BL/6, Balb/c and DBA/2), and 50μg/mouse caused mortality in the majority of CD1 out-bred mice but no/minimal mortality in the other strains. Interestingly, death in CD1 mice was prevented using intravenous immunoglobulin (IVIg) therapy, despite no improvement in anemia. TER119 caused a similar degree of anemia in Fcγchain-/-, FcγIIB-/-, C3-/- and C5-deficient mice as compared to WT mice, suggesting that mechanisms in addition to FcγR/complement-mediated phagocytosis and C5-mediated lysis might be contributing to RBC destruction. Because passive ITP is fully prevented in the Fcγchain-/- mice, the observation that the activating Fc receptors (which all require the Fcγchain to function) may not be the major players in causing TER119-mediated anemia could indicate that mechanisms in addition to FcγR blockade may be responsible for the anti-D-like effect of TER119 in ameliorating this form of murine ITP.

Keyword(s):
• TER119
• RBC
• ITP
• IHA
Pediatric platelet transfusions require smaller volumes of platelet concentrates than what is currently supplied by CBS. A project was conducted to validate the process of taking aliquots from apheresis platelet concentrates (APC) into small volume containers and to assess platelet product quality at the end of storage. Single APC units from consenting donors were collected on CaridianBCT TRIMA and Haemonetics MCS+ automated collection devices. Aliquots were taken into three types of storage containers (Fenwal PL1240 300 mL gas-permeable, Fenwal 4R2014 300 mL gas-impermeable, and 60 mL syringes). Sample aliquots of ~50 mL were taken from 6 Trima units and 6 MCS+ units on day 2, 3, 4 and 5 of storage to represent the time points where hospital customers could take aliquots from an APC unit. The aliquots in PL1240 storage containers were tested for platelet quality on expiry (day 5 post collection) and the other two containers were tested at 4 hours after the aliquot was taken. Data resulting from the validation study indicates that: the process of aliquoting is efficient and can maintain sterility of parent and small volume container platelets; the quality of platelets in small volume containers at 4 h (non-permeable) and 5 d (gas permeable) were generally equivalent to conventional 5 d stored platelets; and the small volume platelets are consistent with the quality standard requirements for pH and sterility. This information should guide blood banks and assist them in making an informed decision about continuing or altering their practice of preparing small volume platelet concentrates for pediatric transfusions.

**Keyword(s):**
- pediatric transfusion
- storage container
- platelet quality
- apheresis platelet
Quality of SAGM, LR RBC Units Washed using the ACP-215 Cell Processor

A Hansen, JP Acker
Canadian Blood Services

Purpose: Washed red blood cell (RBC) units are integral to the Canadian Blood Services (CBS) mandate of providing safe and effective blood products. Under the present system, washed SAGM, LR RBC units have a 24 h post-wash expiry due to open system processing. In addition, the cells are stored in the residual normal saline-dextrose solution used for washing without any preservative. The advantage of the Haemonetics ACP-215 instrument is that it significantly extends storage time for washed units through the use of sterile docking and the addition of a RBC preservative after processing.

Methods: To evaluate the feasibility of introducing the ACP-215 to CBS manufacturing SAGM, LR RBC units were washed at 2 d (n=6), 7 d (n=6), 14 d (n=6), and 21 d (n=6) post collection. Half the units were re-suspended in SAGM and the remaining in AS-3. ATP, supernatant K+, MCV, MCH, MCHC, hematocrit, and hemolysis were measured immediately post-wash and every 7 days post-wash until the unit expiry at 42 d post collection. This data was used to select the optimal conditions for the validation that was performed by measuring 40 units immediately pre-wash at 14 d post collection and at expiry after 7 d of post-wash storage in SAGM.

Results: ACP-215 was able to produce washed RBC units that maintained an average of 97 ± 2.6% of the original hemoglobin resulting in average total hemoglobin of 52 ± 8 g/unit and a hematocrit of 0.55 ± 0.04 L/L. The average hemolysis and supernatant potassium observed after 7 d post-wash storage was 0.27 ± 0.06% and 10.7 ± 1.4 mmol/L respectively.

Conclusions: The results of RBC quality testing performed to examine the effect of pre- and post-wash storage and the post-wash preservation on the in vitro quality of the RBC units indicated that a SAGM, LR RBC unit washed at 14 d post collection using the ACP-215 can be stored in SAGM for 7 d post-wash.

Keyword(s):
• RBC quality
• washed RBC
• haemonetics ACP-215
Novel Mouse Anti-Mouse/Human GPIbα Monoclonal Antibodies: Development and Characterization of New Reagent in Transfusion Medicine

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Background: GPIbα is one of the major target antigens in ITP. However, the mechanism of platelet destruction and the role of IVIG treatment are still not fully understood. Current monoclonal antibodies (mAbs) used in research are generated using traditional methods involving cross-species immunization. These approaches may generate a limited repertoire of mAbs and ITP studies are hampered by the use of xenogeneic antibodies in murine models.

Methods: We developed anti-GPIbα mAbs in GPIbα-/- mice. Platelet binding and specificity were determined by flow cytometry. Antibodies’ effects on platelet function and destruction were measured using aggregometry, intravital microscopy and i.v injection to induce ITP in mice.

Results: Nine mAbs were generated against GPIbα. Three mAbs also bind to human platelets. These mAbs inhibited human platelet aggregation induced by ristocetin and thrombosis formation on collagen coated surface. Some mAbs inhibited mouse platelet adhesion and thrombus formation in vivo. These mAbs induced differing degrees of severity of ITP in mice.

Conclusions: We generated mouse anti-mouse/human GPIbα mAbs. These mAbs had different effects on platelet function and destruction. These mAbs should be very useful reagents for research in transfusion medicine.

Keyword(s):
- ITP
- IVIG
- platelets
- GPIb

Notes
Characterization of Novel Monoclonal Antibodies that Target Murine and Human Beta3 Integrin

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Background: ITP is immune disorder caused by antibodies, a majority against platelets β3 integrin. However, the mechanism of platelet destruction and the role of IVIG treatment are still not fully understood. Current mAbs are generated using traditional methods involving cross-species immunization. These approaches may generate a limited repertoire of mAbs and ITP studies are hampered by the use of xenogeneic antibodies in murine models.

Methods: We developed anti-mouse β3 integrin mAbs in β3-/- mice. Platelet binding and specificity were determined by flow cytometry and western blot. In vitro effects on platelet function were measured using aggregometry. I.v. injections of mAbs were used to induce thrombocytopenia, and the efficacy of IVIG was evaluated.

Results: A total of twelve mAbs were generated. The mAbs were specific for β3 integrin. The anti-PSI domain mAbs recognized linear epitopes. A majority of mAbs cross-reacted with human platelets. Some of them inhibited human platelet aggregation. These antibodies induced differing degrees of severity of ITP, which were variably responsive to IVIG.

Conclusions: We developed mouse anti-mouse/human β3 integrin mAbs, which had different effects on platelet function and destruction. These mAbs should be very useful reagents for research in transfusion medicine.

Keyword(s):
- ITP
- IVIG
- platelets
- beta3 integrin
Anthocyanins Inhibit Platelet Activation in Both Human and Murine Thrombosis Models

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Background: Anthocyanins are major phytochemicals abundant in plant food and have been shown to play a protective role against cardiovascular diseases (CVDs). However, its roles in platelet function and thrombosis are largely unknown.

Methods: Cyaniding-3-gulucoside (Cy-3-g) and delphinidin-3-glucoside (Dp-3-g), the two predominantly bioactive compounds of anthocyanin preparations, were incubated with gel-filtered platelets and platelet rich plasma (PRP) from human and C57BL/6J mice. Platelet aggregation and adhesion were assessed by an aggregometer and perfusion chambers. The effects of anthocyanins on thrombus formation in C57BL/6J mice were assessed using an intravital microscopy thrombosis model.

Results: Both Cy-3-g and Dp-3-g significantly inhibited platelet aggregation induced by collagen and TRAP in gel-filtered platelets, and inhibited aggregation induced by ADP, TRAP and collagen in human and mouse PRP. These inhibitory functions were observed at Cy-3-g and Dp-3-g doses as low as 0.5µM. These compounds also markedly reduced thrombus growth in perfusion chambers at both low and high shear rates. Using intravital microscopy, we further demonstrated that Cy-3-g and Dp-3-g decreased platelet deposition, destabilized thrombi, and prolonged the time required for thrombus formation and vessel occlusion.

Conclusions: Anthocyanin directly inhibited platelet aggregation, and attenuated thrombus growth at both arterial and venous shear stresses. These effects on platelets likely contribute to the protective effects of anthocyanins against thrombosis and CVDs.

Keyword(s):
• anthocyanins
• platelet
• thrombosis
• aggregation

Notes
The Effect of Exogenous Erythropoietin on Hemoglobin Levels in Patients with Baseline Hemoglobin < and ≥ 130 g/L

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Background: At the Vancouver Hospital Blood Utilization Program (VHBUP), the use of exogenous erythropoietin (EPO) in preoperative patients with baseline hemoglobin (Hb) ≥ 130 g/L is at the discretion of the physician. The efficacy of EPO in such patients is unknown. The objective of this study was to compare EPO’s effects on patients with a baseline Hb < 130 g/L to those with ≥ 130 g/L.

Methods: This was a retrospective chart review from January 2008 to June 2009 of patients who received EPO preoperatively for blood conservations. Collected data included the baseline Hb level at time of referral, number of doses of EPO received, and preoperative Hb to assess the change in Hb per dose of EPO.

Results: A total of 279 patients were reviewed of which 221 had documented pre- and post-EPO Hb levels. Patients received a dose of either 20K or 40K depending on weight (< or ≥ 65kg respectively). They received between 1-4 doses of EPO along with supplemental iron (IV and/or PO) unless they refused. EPO was associated with a mean Hb increase of 8.3 g/L per dose (95% CI 7.3-9.2 g/L) amongst all study patients. Patients with a baseline Hb < 130 g/L (n=143) had a mean increase of 9.7 g/L per dose (95% CI 8.5-10.9 g/L). Patients with a baseline Hb ≥ 130 g/L (n=78) had a mean increase of 5.7 g/L per dose (95% CI 4.3-7.1 g/L).

Conclusions: Preoperative EPO administration was associated with an increase in preoperative EPO administration was associated with an increase in preoperative HB in the studied VHBUP patients. The increase was less significant in patients with a baseline HB ≥ 130 g/L. Whether EPO can reduce the risk of allogeneic transfusion in this group of patients should be assessed.

Keyword(s):
• erythropoietin
• blood conservation
• hemoglobin

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Notes
Co-transplantation of _ex vivo_ expanded progenitors with non-expanded (N-E) hematopoietic stem cells improves the recovery of neutrophils while that of platelets remains problematic. The lack of efficacy on platelet recovery could be the result of poor expansion of megakaryocyte progenitors (Mk-P) induced by the cytokine cocktails currently used. Therefore, we recently developed the cocktail OMPC for the expansion of CB Mk-P. Herein, we first investigated whether expansion of cord blood (CB) cells at 39°C was advantageous for platelet recovery, since we previously showed that MH improved the expansion and differentiation of Mk ex vivo. Short-term human platelet (hPLT) recovery was found improved in mice transplanted (TX) with CB cells expanded with OMPC at 39°C, while no significant differences were observed for the long term hPLT recovery. Next, the short-term hPLT potential of CB CD34+ cells expanded with OMPC were compared to that of N-E cells. Only low and insignificant hPLT levels (p>0.05 vs PBS-control group) were detected 4- and 7 days post-TX (PT) with the N-E cells (median < 1.4 hPLT/ul). In contrast, hPLTs were readily detected in the majority of mice injected with OMPC-expanded CB CD34+ cells (9- and 26-fold greater at day-4 and day 7, p<0.05). Conversely, hPLT levels in mice injected with N-E cells became and remained greater past 11 days PT. Similar results were observed with mobilized peripheral blood CD34+ cells. Hence, ex vivo expansion of CD34+ cells in optimized MK conditions can improve their short-term hPLT potential.

**Keyword(s):**
- platelets
- cord blood cells
- transplantation
- CD34+ cells
Development of a Provincial Agglutination Competency Tool

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1Fraser Health, 2Canadian Blood Services, 3BC Provincial Blood Coordinating Office Columbia, 4Interior Health, 5Northern Health

**Purpose:** The Technical Resource Group of the BC Provincial Blood Coordinating Office (PBCO) and Canadian Blood Services (CBS) collaborated to develop tools to standardize grading of tube agglutination in BC laboratories. These tools include standardized provincial technical procedures for Reading and Grading Tube Hemagglutination Reactions, Preparing Reagents and Performing the Agglutination Grading Competency.

**Methods:** To reduce variables, Canadian Blood Services provides R2R2 concentrated cells to hospitals and testing is performed using calibrated pipettes and a five minute room temperature incubation before reading. After initial competency testing and to further reduce variables, Canadian Blood Services provided pre-prepared dilutions for participating hospitals. In addition, some health authorities prepared one 3% dilution of the R2R2 cells to further reduce variables and focus on reaction grading.

**Conclusions:** As tube agglutination techniques in transfusion medicine continue to decrease, reading and grading challenges increase. Performance of tube agglutination competency testing and review of results with participants increases not only competence but awareness of differences that can arise even when as many variables as possible are removed.

**Keyword(s):**
- agglutination
- variables
- standardizing
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Purpose: Resident training in transfusion medicine (TM) in Canada is provided by a collaborative effort between the five university training programs and Canadian Blood Services (CBS). In order to facilitate more consistent training, we organized weekly educational sessions delivered via conference call and webinar.

Methods: Conference calls (Bell Conferencing, Inc) and webinars (GoTo Webinar, Citrix Online) were hosted from the CBS centre in Toronto. A CBS-hosted web page (www.transfusionmedicine.ca) displayed the conference calendar, and lecture slides were available for download. An e-mail survey (SurveyMonkey) allowed us to track our audience and provide feedback to speakers. Attendance was not limited to TM residents. Participants received RCPSC Maintenance of Certification program education credits for attending.

Results: Since the start of academic year 2010, 10 educational sessions were presented. 379 incoming calls were hosted (median 40.5, range 23-53), some of which represented multiple attendees on speakerphone. 344 e-surveys were returned (median 34.5, range 19-51) with average 6% residents, 28% staff physicians, and 56% laboratory staff. The majority were from ON, but all provinces and territories with the exception of NB, YT, NT were represented. Respondents felt the presentations were effective and relevant to their practice/training.

Conclusions: Emerging communications technologies can be leveraged to facilitate education sessions for small numbers of widely dispersed resident trainees. These sessions can be made available to a larger audience such that the entire community benefits.

Keyword(s):
- resident
- education
- information technology
Going Beyond Borders

The Ottawa Hospital

**Purpose:** In July 2010, CBS, North/East Ontario and Nunavut, received a request to provide transfusion support for a patient with aplastic anemia and anti-AnWj. The AnWj- phenotype is very rare; but, patients with anti-AnWj may also receive cells of the In(Lu) phenotype. The patient was expected to need transfusion support up to time of transplant and until engraftment. Patient’s transplant failed to engraft fully and on-going regular transfusions were needed.

**Methods:** A rare donor unit search was initiated. No AnWj- units/donors were found at CBS or Hema-Quebec and only limited In(Lu) units/donors were available. These resources were quickly exhausted and the search was extended to the U.S. Between August-December 2010 the American Rare Donor Program provided a total of 11 units, fresh and frozen In(Lu) and AnWj-. As the holiday season approached, European blood centres were contacted to search for compatible units/donors. The search identified units in the UK and France. The UK shipped 2 thawed units with 7 day expiry. No additional units were needed as patient expired shortly thereafter.

**Results:** International collaboration enabled transfusion support for a prolonged period of time for a patient with a rare antibody. The treatment plan was modified for the month of December, to ensure patient treatment was not compromised over the holidays. Going outside the country required more time, foresight and planning to mitigate issues such as having shipments bumped off flights, units breaking in transit and product life span. Contrary to the North American standard of 24 hours, the UK and France have 7 day expiry post thaw of frozen product which was beneficial for overseas transfers.

**Conclusion:** Sharing of rare blood is crucial to the survival of these challenging patients. International shipping of blood products poses unique challenges.

**Keyword(s):**
- AnWj - rare donor
- In(Lu) - international
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¹Centre for Blood Technology Operations at Dalhousie University, ²Canadian Blood Services

**Background:** Canadian Blood Services (CBS) has developed a plan to consolidate production and support functions for the Maritime region in Dartmouth, NS. Once consolidated, blood products for New Brunswick hospitals will be distributed from the Dartmouth production facility either directly or via a stock holding unit located in Saint John. CBS has committed to a standard of service under the new network that is at least as good as that of the existing network.

**Purpose:** To evaluate and compare expected levels of customer service for NB hospitals under the current and proposed distribution networks.

**Methods:** A discrete event simulation model of the Saint John centre was developed. The simulation model represents processes of inventory management (collections, testing, end-labelling), distribution (order arrivals, order completion, dispatch) and transport (travel time, weather delays, road closures). An MS-Access database tracks the location and status of components in the model.

**Results:** System performance metrics (order fill time and outdate rates) were recorded for both the current and proposed distribution networks using the simulation model. A comparison of means tests (α = 0.05) indicates that the proposed network has performance metrics that meet, or exceed, those of the existing network.

**Conclusions:** Levels of customer service experienced by NB hospitals will not be degraded by consolidation of production and support facilities in Dartmouth, NS.

**Keyword(s):**
- inventory management
- distribution
- logistics
Purpose: In June 2010, G8 and G20 Summits were held in Huntsville and Toronto, Ontario. CBS developed and implemented plans to limit disruption of service to its clients during these events.

Methods: Action plans were built on the foundation of emergency plans developed to deal with service disruptions resulting from Pandemic Influenza. Preparations focused on two major components: internal and external stakeholders. Internal stakeholders were defined as the staff and departments who comprise Central Ontario Region. The LERT (Local Emergency Response Team) in each site assisted departments in performing gap analyses of anticipated impacts and in the planning to ensure CBS could continue to meet hospital demand. Contact was made with external stakeholders including all hospitals served by CBS, the Ontario Ministry of Health Emergency Management Services, Health Canada and local police services.

Results: Making connections with these organizations was critical in moving product during the period of riots, road closures and civil unrest. NERT (National Emergency Response Team) members were kept informed of developments and were available for assistance if required.

Conclusions: Lessons learned included the realization that early planning and outreach to outside agencies were vital in dealing with disruptions of this nature. Maintaining these relations should be a priority of any agency providing an essential health service. Lessons were shared with other CBS sites so that all may benefit from local experiences.
**Shannon Selin, Stephanie Bowen, Susanna Darnel* 
**BC Provincial Blood Coordinating Office**

**Purpose:** The National Plan for the Management of Shortages of Labile Blood Components (2009) recommends the establishment of Emergency Blood Management Committees (EBMCs) in each province/territory and hospital/ regional health authority to serve as communication conduits and to oversee the implementation of local blood shortage management plans. These committees may need to meet at short notice in an emergency, under circumstances in which local communications infrastructure may not be fully functioning. The British Columbia Provincial Blood Coordinating Office (PBCO) has developed a procedure for convening the BC EBMC that can be used at any time, by any member, regardless of whether the PBCO is open, the Chair is available, or phone lines are functioning in the earthquake-prone BC Lower Mainland.

**Methods:** The procedure – which can be initiated by any BC EBMC member – relies on a wireless incident command broadcast messaging system. The system immediately opens a teleconference line and broadcasts instructions on how to join the teleconference to all BC EBMC members at their office, home and mobile telephone and email addresses. If phone lines are not working in the Lower Mainland, a backup procedure enables participants to send a group email (to a listserv) to start a teleconference using a line outside the Lower Mainland.

**Results:** The procedure was tested in October 2010 and was used to convene a BC EBMC meeting during a platelet shortage in November 2010. Most members were on the line in under 30 minutes.

**Conclusions:** A procedure using a broadcast messaging system is a feasible way to quickly convene an EBMC.

**Keyword(s):**
- blood shortage
- Emergency Blood Management Committee

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**Notes**
The IMT: The Nova Scotia Approach to Enhance Inventory Management During Blood Shortages

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Background: Blood Contingency planning in Canada has been evolving nationally and provincially. In September 2009 the National Plan for the Management of Shortages of Labile Blood Components was released. In April 2010, the NSPBCP released the N.S. Provincial Blood Contingency Plan. The plans are structured around phases of inventory described as Green, Amber, Red and Recovery. Phases are interpreted as a function of the number of units equated to “days on hand”. In times of shortage or imminent threats, CBS requires hospital specific inventory levels. In N.S. an Inventory Management Tool, known as the IMT was created to collect inventory information.

Approach: Blood inventory is a national commodity; in creating the IMT; N.S. chose to focus on creating a metric that neutralizes facility specific inventory levels and relates it to service using the same metric as National” days on hand”. The IMT itself is a series of Excel spreadsheets designed to summarize N.S. hospital inventory, which captures nine District Health Authorities (DHAs) and a Children/ Women’s specialty centre (IWK); CBS-Halifax local inventory and CBS –National inventory for RBCs and Platelets. DHAs/IWK are provided with Excel spreadsheets specific to their organization. The sheets are organized by Group and Rh, categories include available inventory, units crossmatched and units outdating within “x” days. Facility optimum inventory is entered on the sheet; the data entered becomes a function of optimum and is translated to “days on hand”.

Results: Reviewing a common metric promotes, transparent provincial Inventory Management decision making during shortages/threats to the blood supply.

Keyword(s):
- inventory
- shortage
- management
- metric

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Notes
Hendry, Julia*; Gosling, Janice; Shabani-Rad, Meer-Taher
CLS/University of Calgary

Purpose: Application of the utilization inventory index to hospital red cell stocks to determine if red cells stocks could be decreased in an effort to provide fresher blood for transfusion.

Method: Red cell utilization data was analyzed for the period January 2009 to September 2010 for our 4 urban hospitals. Utilization was broken down by ABO/Rh and site of transfusion. Based on the modeling from our Blood Utilization Management Plan, new regional and site specific optimal inventory ranges and triggers for green, red and amber inventory management phases were set. The inventory phase triggers were set using combined days on hand of CBS local inventory and CLS regional inventory. Regional and site specific inventory ranges were set based on 7 to 9 days of average utilization (inventory index). These new inventory ranges were compared to the previous levels and evaluated to ensure suitable inventory would be available at each site to manage bleeding emergencies.

Results: It was apparent we were overstocking red cells. An independent analysis of the age of blood at the time of transfusion showed a disproportionately large number of units were being transfused in their fifth and sixth week of shelf life. The following adjustments to the inventory ranges were made. Amounts of B negative, AB positive, and AB negative were increased to allow a minimal stock of these groups at all three adult sites. Our pediatric facility uses only 5% of all red cells transfused in our region. The inventory ranges for our Children's Hospital could not be based on utilization data as they are a level 1 trauma facility and thus must be stocked at levels higher than their utilization data indicates. Regional inventory ranges were adjusted to reflect the increased red cell stock at the Children's Hospital.

Conclusion: Basing hospital red cell inventories on the utilization inventory index may decrease the number of red cells stocked and provide fresher red cells for transfusion; however implementation requires a well coordinated effort with the local CBS to ensure adequate red cell stocks and prevent increased age of red cells at time of issue from CBS. The use of utilization inventory index is only applicable to group O+, O-, A+, A-, and B+ red cells stocked at medium to large adult facilities.

Keyword(s):
- red cell utilization
- red cell inventory

Notes
Stephanie Watton*, Transfusion Practice Coordinator, Nova Scotia Provincial Blood Coordinating Program (NSPBCP); Marina Hamilton, Program Manager, NSPBCP

**Background:** The NSPBCP in collaboration with the Nova Scotia Nurses Transfusion Practice Working Group (NSNTPWG) recently created a “Toolkit for the Administration of Blood Components and Blood Products”. This toolkit provides a template for hospital sites in Nova Scotia for use in the development of facility specific policies for administration of blood components and blood products. As part of the template package, a transfusion safety checklist was developed based on the awareness of the critical role that positive patient identification plays in the transfusion process. Serious/fatal transfusion reactions have occurred related to errors in patient identification. This bedside checklist provides a step-by-step reference tool for the transfusionist to support documentation and ensure the critical aspects of blood administration have been performed. This tool supports CSA Standards Z902-10 for blood and blood components and ensures safe and best practice.

**Method:** The NSPBCP performed a literature review to consider existing patient safety checklists. Resources such as Surgical Safety checklists have been developed by organizations like the Canadian Patient Safety Institute, and the World Health Organization (WHO). Patient safety tools such as the transfusion safety checklist, supports one of the goals of the NSPBCP’s strategic plan; to reduce preventable adverse events.

**Results:** Implementation of this checklist tool throughout Nova Scotia hospitals will ensure safe transfusion practices and decrease the potential for error. Utilizing the checklist at the bedside will direct the transfusionist in the required steps for blood administration and will support safe patient care and compliance with blood safety standards.

**Keyword(s):**
- transfusion
- patient safety
- checklist
- blood administration
Provincial Standard for the Investigation of Adverse Transfusion Reactions in Nova Scotia

Stephanie Watton*, Transfusion Practice Coordinator, Nova Scotia Provincial Blood Coordinating Program (NSPBCP); Wendy Varrence, Lab Standards Coordinator* (NSPBCP); Marina Hamilton, Program Manager, NSPBCP

Background: To reduce preventable adverse events is an identified strategic goal of the NSPBCP. The first step in order to achieve this is to accurately identify the current risks to patients. The ability to accurately classify reactions is based on the results of the investigation performed. The NSPCBP identified that investigations of adverse reactions were conducted differently throughout the province and a standardized approach to investigating adverse transfusion reactions was needed. Concurrently, “Guidelines for the Investigation of Suspected Transfusion Transmitted Bacterial Contamination”, was published. This guideline advised on the investigation of suspected bacterial contamination reactions but also identified a gap in the investigation of other suspected transfusion reactions.

Method: A literature review was completed and applicable blood and blood component standards such as AABB and CSA Standards Z902 were reviewed. The provincial standard was created in collaboration with the NSPBCP’s adverse reaction investigation working group consisting of provincial representation from nursing, laboratory technologists, laboratory medical directors and Canadian Blood Services, and received support for implementation from NSPBCP stakeholders.

Results: The provincial investigation standard assists healthcare professionals in gathering evidence to classify adverse transfusion reactions appropriately and includes an algorithm to support clinical identification as well as a laboratory investigation chart which supports the recommended laboratory and clinical testing required for classification.

Conclusion: Accurate classification of transfusion reactions promotes patient safety by encouraging timely and appropriate treatment of the patient. Implementation of this provincial standard promotes a consistent approach to adverse reaction investigation throughout Nova Scotia.

Keyword(s):
- algorithm
- investigation
- adverse reactions
The Value of Prescribing a Transient Course of Oral Iron and Vitamins as a Preoperative Blood Conservation Strategy

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Purpose: A suboptimal hemoglobin level increases transfusion risk for patients scheduled to undergo high blood loss surgical procedures such as total joint replacement and radical prostatectomy. The purpose of this study is to assess the value of prescribing a short course of oral iron and vitamins to non iron deficient patients ineligible for other pre-operative blood conservation strategies such as erythropoietin therapy, IV iron or autologous blood donation.

Methods: A retrospective review was conducted on ninety-nine pre-operative patients referred to the Ottawa Hospital blood conservation program who received oral iron, folic acid and vitamin B12 as the primary hemoglobin (Hgb.) optimization strategy.

Results: Overall, the majority of cases experienced a hemoglobin increment (77%) prior to surgery. Average duration of treatment was nineteen days. A stronger Hgb. response was noted among the prostate group as compared to orthopedics (88% vs. 78%). Forty-two percent of the prostate group demonstrated robust Hgb. increments ranging from 10-16g/L., whereas only 20% of the orthopedic group showed a similar response. There was good adherence to therapy and no adverse effects were reported.

Conclusion: A short course of oral iron, folic acid and vitamin B12 appeared to be a safe and well tolerated strategy to attempt to optimize pre-op hemoglobin levels for surgical patients ineligible to receive other pre-operative blood conservation modalities.

Keyword(s):
- hemoglobin optimization
- blood conservation
Education Materials for Patients and Physicians Regarding Transplantation Adverse Events (TAEs) In Nova Scotia

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Nova Scotia Provincial Blood Coordinating Program

Purpose: As a result of an environmental scan and physician survey conducted in Nova Scotia surrounding TAEs, a number of recommendations were brought forth. Two of these recommendations included developing education materials surrounding the identification, diagnosis, and reporting of TAEs to the appropriate bodies provincially and nationally, specifically related to TAEs following tissue transplants.

Methods: An environmental scan was conducted to identify key stakeholders in the organ and tissue donation and transplantation community in Nova Scotia and to identify the current reporting structures that exist. Additionally, a physician survey was conducted to determine physician knowledge surrounding the Canadian Standards Association Z900.1-03 Cells, Tissues, and Organs for Transplantation and Assisted Reproduction, specifically related to the reporting of TAEs. From the results of the scan and the survey, a number of recommendations were developed, including the need for family physician and post-operative transplant patient educational needs surrounding the identification, diagnosis, and reporting of TAEs. Education materials entitled “Identifying and Reporting a Transplantation Adverse Event after Tissue Transplantation” developed for family medicine practitioners and “Tissue Transplant: A Patient Guide” developed for use by patients.

Results: Education materials that will assist transplant patients in determining when to seek medical advice and will assist family physicians in identifying and correctly reporting TAEs in their post-operative transplant patients.

Conclusions: While the standards exist surrounding the reporting of TAEs, it is important to ensure that end users of tissue products, care providers, and patients are aware of what, how, and to whom to report.

Keyword(s):
- transplant
- adverse event
- patient
- family medicine practitioner

Notes

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Patient Pamphlet Evaluation Survey

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Ontario Regional Blood Coordinating Network (ORBCoN)

Purpose: To evaluate an educational resource “Blood Transfusion: A Patient’s Perspective” launched by ORBCoN in September 2008.

Methods: Two evaluation strategies were used to identify any required changes. Ten (10) patients at each of 9 hospitals, as well as health care professionals (HCP) at all hospitals with transfusion services were invited to provide feedback. Patients and HCP completed a written survey with the following categories: Length, Comprehension, Content, Applicability and Format of the pamphlet.

Results: 69.4% (93/134) completed responses were received from surveys sent to HCP; 59% of responding sites use the pamphlet. Patient responses analyzed to date show 73% answered that the pamphlet provided a sufficient level of information.

Conclusions: This evaluation feedback reveals both specific changes that can be made for the second edition, and which sections need to be kept intact. Evaluation also served as an awareness strategy since some HCP were unaware the pamphlet existed. Alternate uses for the pamphlet were also reported.

Keyword(s):
- patient
- transfusion
- education
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The Ottawa Hospital

**Purpose:** Investigation of 19 plasma donors was requested after report of a possible TRALI during a plasma exchange in a patient with TTP. As all donors were males, it was felt to be unlikely that antibody-mediated TRALI could explain the patient’s symptoms although all clinical criteria for TRALI were met.

**Methods:** The implicated donors were contacted to provide samples for HLA testing. 15 of the 19 donors submitted samples. Testing for HLA type and anti-HLA antibodies was done at the CBS platelet immunology lab using Luminex technology. Antibody positive donors were then contacted to ask about transfusion history. HNA testing was not available at the time of the investigation.

**Results:** Testing showed that three of the men had HLA antibodies that did not correlate to the patient’s HLA type. They were unlikely to have caused antibody-mediated TRALI. One donor had a strong anti-HLA antibody directed towards the cognate antigen in the patient. (See Figure 1). None of the donors had a history of transfusion, major surgery or any kind of tissue transplantation.

**Figure 1:**

<table>
<thead>
<tr>
<th>Patient Test Results - HLA Typing:</th>
</tr>
</thead>
<tbody>
<tr>
<td>A2, Cw7, Cw8, DR7, <strong>DR17</strong>, DR52, DR53, DQ2, DQ9</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Donor Test Results - HLA Antibody Specificity:</th>
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<tbody>
<tr>
<td>Donor 1</td>
</tr>
<tr>
<td>Donor 2</td>
</tr>
<tr>
<td>Donor 3</td>
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<td>Donor 4</td>
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</table>

**Conclusions:** Clinical and immunologic criteria for antibody-mediated TRALI were demonstrated in this case, highlighting the importance of maintaining a high index of suspicion for TRALI regardless of donors involved and donation history.

**Keyword(s):**
- TRALI
- HLA
- male plasma
- ATR
Blood Warmer Use in Massive Transfusion in Nova Scotia

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Nova Scotia Provincial Blood Coordinating Program (NSPBCP)

**Purpose:** The Guideline for Massive Transfusion in Nova Scotia was disseminated in December, 2010. In patients who are massively bleeding, it is recognized that the delivery of cold blood components contributes to hypothermia and adverse patient outcomes. Therefore, the guideline advises warming RBCs and plasma in this setting. The purpose of this study is to determine the current practice regarding the warming of blood components administered in a massive transfusion setting within Nova Scotia.

**Method:** A questionnaire was distributed to nurses where massively bleeding patients are typically managed - ORs, ICUs and emergency departments. The survey was anonymous except for the facility and specialty. The survey will also obtain information on specific blood warming devices employed as well as frequency, indication and setting of use.

**Results:** The survey is ongoing and results are pending. The results will:
1. report on the availability of blood warmers, and their use in a massive transfusion setting as reported by the survey participants.
2. describe if blood components are not warmed in massive transfusion, why this is occurring

**Conclusion:** The results may identify educational gaps and equipment difficulties thus providing opportunity for educational sessions regarding the warming of blood components in massive transfusion settings. The results of the survey may provide product recommendations for hospitals to consider when purchasing blood warming equipment.

**Keyword(s):**
- massive transfusion
- blood warmers
- survey of current practice in NS
**Efficient Life-Span of Red Cells Lost on the Blood Bank Shelves**

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CLS/University of Calgary*

**Background & Purpose:** Utilization of fresh blood has recently been focus of multiple clinical studies. Limitations in supplement of fresh blood have raised serious concerns among blood suppliers and transfusion services. This study was designed to review the age of red cells at three levels of blood system; regional CBS, hospital inventory and transfused red cells.

**Method:** Age of red cells were recorded in a midweek (June 2010) at three levels of “red cells within regional hospitals inventories” and “red cells transfused to the patients”. Then the red cells were categorized in seven groups based on weekly shelf-life as follows: week 1 (<7 days), week 2 (8-14 days), week 3 (15-21 days), week 4 (22-28 days), week 5 (29-35 days) and week 7 (36-42 days).

**Results:** The age of red cells issued by regional CBS were in range of one to four weeks; week 1 (22%), week 2 (13%), week 3 (53%) and week 4 (12%). The age of red cells within the hospital inventories were between week 1 to week 6; week 2 (2%), week 3 (16%), week 4 (37%), week 5 (27%) and week 6 (17%).

**Conclusion:** A significant proportion of transfused red cells were within week 5 & 6 of shelf-age (44%). The transfused red cells appropriately reflect similar shelf-age of red cells within the hospitals inventories. The presence of relatively old blood within the hospitals inventories. The presence of relatively old blood within regional CBS and hospital inventories at the time of study indicates inappropriate high levels of red cell stocks at both sides. Since the high incidence of adverse reactions among transfused patients with old blood have been shown by multiple studies, the significance of this finding is remarkable. Therefore the optimizations of regional inventories by appropriate decrease in the number of red cells for the transfusion. This modification may eventually result in better patient care, less transfusion associated adverse complications as well as correlated financial burden on health care system.

**Keyword(s):**
- red cell shelf-age
- red cell inventory

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**Notes**

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Analysis of Laboratory Information Flow for Recipient Transfusion Notification at Capital District Health Authority (CDHA)

Dalhousie University; Capital District Health Authority

Background: The Canadian Standards Association (CSA) [Z902-10] 11.2.2 indicates that “A procedure shall be in place to ensure that recipients of blood, blood components, and blood products receive notification of the transfusion in writing.” Therefore, we analysed the information flow in our laboratory information system to assess for gaps in doing this electronically.

Methods: In the Fall of 2010, interviews were conducted by Health Informatics students at Dalhousie University with the interface and database systems analysts of the Pathology Informatics Group at CDHA. Flow of information was qualitatively examined.

Results and Conclusions: The generation of patient notification of transfusion is a complex task that requires significant coordination of the hospital information systems. In order to automatically generate letters using patient addresses, the address fields within the admission/discharge system (Mckesson STAR) need to be up-to-date, but this is not often double-checked at the hospital registration desk and updated, and there are confidentiality issues if mail is lost. In addition, the cost of stamps and paper becomes prohibitive over time. If the result is to be sent to a family physician, there are limitations, as not all patients have family physicians. Also, family physician details in STAR do not cross the interface into the laboratory information system in most cases. If the transfusion field is to be populated in a results portal electronically, the current HL7-v2x standard as well as the receiving system may not be able to accommodate this. Given the above considerations, the only available method for delivering the transfusion information with high fidelity to the patient is via manual mailout.

Keyword(s):
- informatics
- transfusion
- notification
- CSA
Optimizing the Use of IVIG in Nova Scotia Using a Request Approval Process

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**Purpose:** In an effort to optimize the appropriate use of IVIG the Atlantic Collaborative IVIG Utilization Working Group (ACIUWG) developed an IVIG request approval system in 2009/10. Nova Scotia implemented the process in November 2009.

**Methods:** Through this process first time or initial requests for IVIG are reviewed to determine if the indication, as well as the dosing, frequency and duration of treatment meet the guidelines for its use. In the event of an incongruity, the ordering physician is contacted and discussion ensues regarding the variation. If the ordering physician continues to feel that a given case merits a change from the guidelines, he or she is asked to discuss the case with a consultant with the relevant clinical expertise. The pathway thus taken by the request is allocated a number representing the route it took for its approval. These pathway numbers are recorded and submitted for each and every new request of IVIG. Currently the project addresses the requests for IVIG for neurological and hematological indications, the two largest areas of adult use. The neurological and hematological guidelines have been developed by the Atlantic IVIG Clinical Experts Working Group and are based on those published by the National Advisory Committee on Blood and Blood Products in Transfusion Medicine Reviews in 2007.

**Results:** There were 46 initial requests in Nova Scotia during the period of November 2009 to March 2010. Thirty nine requests were for hematological and neurological indications and met the guidelines upon initial submission. Seven requests were not applicable to this process as they were for indications not listed in the guidelines.

**Conclusions:** The implementation of the Request Approval Process has been effective in supporting the appropriate use of IVIG in hematological and neurological conditions in the adult population. It is recommended that the adult request approval process be expanded beyond neurological and hematological conditions to include immunological conditions and solid organ transplantation. It is also recommended that the Request Approval Process be implemented in the Pediatric cohort.

**Keyword(s):**
- utilization
- IVIG

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Notes
Jack Michaan, Cheryl Lewis*, Jimmy Chan, Jenn Danielson  
BC Provincial Blood Coordinating Office  

**Purpose:** There are many questions associated to utilization of blood products that could be answered with access to coded data integrated from multiple sources. The Central Transfusion Registry (CTR) contains detailed information about all hospital blood use in BC since April 1999. The RBC transfusion data, however, lacks information regarding reason for transfusion and other important data elements.  

**Methods:** In order to provide more meaningful RBC utilization information, a strategy is underway to link the CTR to other provincial registries in BC. A linkage with the Cardiac Services BC’s Registry has been completed to increase the breadth of information on RBC transfusion for cardiac procedures. Such initiatives present methodology challenges and require that patient’s personal identifiers are automatically and accurately matched between data sets.  

**Results:** Patients existing in both registries were matched and their associated data elements integrated to increase the breadth of information on RBC use and cardiac procedures.  

**Conclusion:** With the aging population increasing pressure to the Canadian blood supply¹ there will be a greater need for utilization information in order to more efficiently manage this valuable resource. Integrating centralized data sources provide an effective way to provide such information.  


**Keyword(s):**  
- red blood cell  
- utilization  
- data linkage
T.Cameron*; W.Owens; H.Nesrallah; K.Gagliardi; S.Anderson; L.Young; D.Lauzon; T.Thompson; S.Cope Ontario Regional Blood Coordinating Network (ORBCoN)

Background: Audits can help identify instances of non-compliance which can lead to potential errors. Identifying potential sources of error can aid in developing and implementing corrective actions to prevent errors before they occur. Hospitals are encouraged to perform audits of their process for transfusion to meet accreditation and improve patient safety.

Purpose: An online audit tool, available to Ontario hospitals was developed to collect data and identify how well hospitals meet current standards for the safe administration of blood components at the bedside.

Method: An ORBCoN working group of Ontario Healthcare Professionals designed an audit of blood administration based on the current CSA and CSTM Standards.

Results: Seven hospitals piloted the program to verify the tool met expectations. Ontario hospitals (158) were then invited to collect data over two months.

Conclusion: Use of the ready made audit form and online tool will facilitate hospitals performing regular audits of their process for the administration of blood, and promote this activity in a standardized manner. Regular performance of audits helps hospitals comply with accreditation and leads to improved patient safety.

Keyword(s):
- audit
- blood component
- standards
- errors

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Notes

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Changes in Consumption of Red Cells (RBC), Frozen Plasma (FP) and Platelets (PLT) in Ontario 2000-2010

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Background: Blood utilization, inventory management measures and transfusion education have been supported by the Provincial MOHLTC over the last 8 years. This study was undertaken to ascertain if there was any associated change in Provincial component consumption.

Methods: Data on issues (reflecting consumption) of RBC, FP and PLT by CBS to Ontario and non-Ontario hospitals for the fiscal years 2000-01 to 2009-10 were obtained from CBS. The issues of the 3 components for Ontario (ON), CBS as a whole (CBS), and the rest of Canada served by CBS (ROC) for each year were calculated as a % of the base year 2000-01. The increments in issues of the 3 components per 1000 population year-over-year were also calculated.

Results: Over the period 2000-01 to 2007-08 (RBC, PLT) and to 2006-07 (FP) consumption increased in ON, CBS and ROC at a similar rate. From 2007-8 to 2009-10 RBC and PLT consumption has declined each year in ON and risen correspondingly in ROC. FP consumption has declined each year in ON from 2006-07 and consumption in ROC and issues by CBS have declined since 2008-09. Calculation of annual increments in consumption in ON of all 3 components/1000 population confirms the decline in consumption over the last few years.

Conclusion: The recent blood utilization and inventory management strategies promoted by the ON MOHLTC are temporally associated with decline in issues and, by inference, transfusion of blood components in Ontario over the last few years. It is tempting to speculate that the decline is a consequence of these measures. The national decline in FP use in 2009-10 may be a consequence of the availability of Prothrombin Complex concentrates.

Keyword(s):
- transfusion
- red cells
- frozen plasma
- platelets

Notes
Utilization Rate Of Five-Day Thawed Plasma Compared To 24-Hour Thawed Plasma

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Background: A previous study at Capital District Health Authority (CDHA) demonstrated that, when compared to plasma thawed for 24 hours, plasma thawed and stored at 1-6oC for 5 days contains levels of labile coagulation factors that are above that required for hemostasis (0.5 U/mL), with no increase in the risk of bacterial contamination.

Method: This study compared the rate of plasma utilization six months pre- and six months post-implementation of 5-day thawed plasma at CDHA in January 2010.

Results: The total wastage rate for fresh frozen plasma (FFP) and frozen plasma (FP) combined decreased from 15.9% (286/1799 units) pre-intervention using 24-hour thawed plasma to 7.16% (88/1229 units) post-intervention using 5-day thawed plasma. There was no increase in plasma utilization or adverse events.

Conclusion: A policy of using 5-day thawed plasma is a safe alternative to 24-hour thawed plasma and leads to reduced product wastage rates, no evidence of increased plasma usage, and no increase in adverse patient outcomes.

Keyword(s):
- plasma
- 24-hour thawed plasma
- 5-day thawed plasma
Frozen Plasma (FP): Red Cell (RBC) Consumption Ratio as an Indicator for Audit

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Purpose: FP: RBC ratios have been used to compare national transfusion practices. We have applied this ratio to individual hospitals as a possible indicator for audit.

Methods: Issues of FP (excluding cryosupernatant) and RBC to Ontario hospitals for FY 2008-9 (CBS) and 2008 Active Treatment Bed numbers (ATB) for 2008 (MOHLTC) were obtained. FP: RBC (as a %) and FP/ATB/year were calculated.

Results: Hospitals were stratified in 3 groups – University-affiliated (U-a), Large Community >200 ATB (LCH) and Medium-sized Community 100-200 ATB (MCH) hospitals. Results are summarized in the table. The 2 indicators correlate well, suggesting both provide a similar measure of practice.

Conclusion: The wide variation within and between hospital groups suggests hospitals at the high end of their group should consider audit of FP use.

<table>
<thead>
<tr>
<th>Hosp</th>
<th>FP/RBC</th>
<th>FP/ATB</th>
<th>r</th>
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<tbody>
<tr>
<td>U-a</td>
<td>11-71.4</td>
<td>2.2-19.1</td>
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<tr>
<td>LCH</td>
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<td>2.4-7.8</td>
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Keyword(s):
- plasma
- audit
- transfusion
Producing Platelet Concentrates Made By Pooling Only Four Buffy Coats with the New Atreus/OrbiSac System: An Evaluation

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Purpose of the investigation: Héma-Québec has recently introduced the preparation of buffy coat-derived platelets with the new Atreus/OrbiSac system (CaridianBCT). With this system, platelet concentrates (PCs) are obtained by pooling 5 buffy coats (BCs). In this work, we have evaluated the possibility of pooling only 4 BCs instead of 5.

Methods: Whole blood (WB) (450 mL) was rapidly chilled and stored at 20-24°C using cooling plates. WB units were processed either within 10 hours or at least 16 hours after collection. PCs were prepared from either 4 or 5 BCs (control) after 3-5 hrs or 20-24 hrs of holding time. PCs, RBC and plasma units were stored at 20-24°C, 4°C and 20°C, respectively. In vitro parameters of blood products were compared.

Results: Instrument settings for the 4-BC pooling process increased RBC loss, resulting in a decreased level of haemoglobin compared to control RBC units (46 ± 5g vs. 50 ± 6g; p<0.05). PCs prepared by 4-BC pooling contained significantly fewer platelets than control PCs (3.0 ± 0.7 vs. 3.9 ± 0.5 x 10¹¹/unit; p<0.05). Nevertheless, blood products processed using 4-BC pooling fulfilled the CSA’s requirements.

Conclusions: Processing of fresh WB using the 4-BC pooling method might lead to inadequate products but longer holding time always resulted in good quality products. Additional work is required to optimize platelet and RBC recoveries.

Keyword(s):
- blood product
- process optimization
- buffy coat
- platelet concentrate

Notes
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**Purpose of the investigation:** Since October 2010, whole blood-derived platelet concentrates delivered by Héma-Québec are prepared by the buffy coat method using the new Atreus/Orbisac system (CaridianBCT). The study was designed to evaluate the compatibility of the Atreus/Orbisac bags with three transfusion sets widely used by hospitals in the province of Québec.

**Methods:** Transfusion sets were obtained from Baxter, Hospira and Alaris Medical Systems. Forces applied for spiking and un-spiking were measured with an in-house apparatus. Assays were carried out with bags filled with water having volumes and temperatures identical to those of blood products. Pressure tests were carried out to check leaks from spike connections. Hospital staff also performed a blind evaluation of the ease for spiking and un-spiking component bags.

**Results:** Differences in spiking forces were observed between transfusion sets with forces of $91.7 \pm 15.8$, $86.9 \pm 15.3$ and $71.5 \pm 12.9$ N for the Baxter, Alaris Medical Systems and Hospira Inc. sets, respectively. Bag septum was always perforated and no leakage occurred. Average spiking and un-spiking forces were similar to our current bags. The similarity between test and control bags in spiking and un-spiking procedures was also confirmed by the hospital staff.

**Conclusion:** This study confirms the good compatibility of the Atreus/Orbisac component bags with the three main transfusion sets used by our hospitals. These findings have greatly facilitated the introduction of the Atreus/OrbiSac component bags.

**Keyword(s):**
- blood product
- transfusion set
- validation study
- process optimization
Platelet Concentrates: How Many Red Blood Cells Are Still in the Bag?

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**Purpose of the investigation:** In the province of Quebec, platelet concentrates (PCs) are prepared either by single-donor apheresis or, more recently, by the pooling of buffy coats (BC). In this work, we have investigated the amount of residual red blood cells (rRBCs) in these PCs.

**Methods:** 150 PCs of each type were sampled on day 1. Samples were stained with anti-CD41 (platelet) and anti-CD235a (rRBCs) and analyzed by flow cytometry. rRBCs (CD235a+ cells) were next analyzed by size and granularity. The assay linearity was tested and validated with a PC spiked with a predetermined number of RBCs. rRBC counts were also compared to manual Neubauer chamber counts.

**Results:** The assay has a linear range of 300 to 10,000 rRBCs/µL ($R^2=0.998$). Average rRBCs counts of apheresis-derived PCs were $2.9 \times 10^7 \pm 2.2 \times 10^7$ rRBCs/unit (range: $0.4 \times 10^7$ to $16.7 \times 10^7$ rRBCs/unit; median: $2.5 \times 10^7$ rRBCs/unit). For BC-derived PCs, the average counts were $12.1 \times 10^7 \pm 5.4 \times 10^7$ rRBCs/unit (range: $1.1 \times 10^7$ to $31.6 \times 10^7$ rRBCs/unit; median: $11.2 \times 10^7$ rRBCs/unit). Blood product volumes were lower for apheresis-derived PCs than BC-derived PCs ($223 \pm 17$ mL vs. $349 \pm 16$ mL).

**Conclusions:** rRBCs counts in PCs satisfy AABB Standards ($<3 \times 10^{10}$ rRBCs/unit) and the European Community Recommendations ($<1.3 \times 10^9$ rRBCs/unit) which have been defined to prevent RBC alloimmunization.

**Keyword(s):**
- platelet concentrates
- blood product
- alloimmunization
- quality contol
Purpose of the investigation: Héma-Québec collects about 225,000 blood donations yearly. The goal of this study was to evaluate the workflow and turnaround time of the Ultrio (HIV-1, HCV, HBV) and WNV assays on the Procleix Tigris system according to 3 WNV seasonal scenarios (winter, summer and summer with individuals testing).

Methods: One Tigris and one pooling system were installed and validated. About 0.08% of plasma samples were spiked with a positive sample to mimic a typical incidence rate. Samples were analyzed with the Tigris system in pools or individually, based on three seasonal WNV scenarios (#1: Ultrio=5431, WNV=544; #2: Ultrio=5431 WNV=5431; #3: Ultrio=5432, WNV=2716 + 2716 (individual)). Work was performed on 7-hour shifts with 20-minute breaks and a 1-hour lunch.

Results: Turnaround times were 11.2 ± 2.0, 11.8 ± 0.7 and 14.4 ± 1.5 hours for scenarios 1 to 3, respectively. Operator’s hands-on times were respectively 3.0 ± 0.8, 4.1 ± 0.6 and 5.1 ± 1.1 hours for these 3 scenarios. Starting analysis at 7 a.m., the first set of complete results was released for revision at 1:58 p.m., 2:31 p.m. or 7:41 p.m. for scenarios 1 to 3. For all scenarios, laboratory staffing needs were 2 operators for the day shift and one for the night shift.

Conclusion: This work shows that turnaround times can be successfully achieved with a minimal instrument set-up and a limited laboratory staff.

Keyword(s):
• process analysis
• testing
• operational trial
• automation
Evaluation of the Need and Cost of RBC Genotyping in a Teaching Hospital

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Background: RBC genotyping may help identify optimally matched RBC for a patient while saving several hours of work on the bench in transfusion laboratory. Unfortunately many hospitals do not have access to genotyping in their laboratory and sending samples out may be associated with a significant cost. In this study we evaluated the need for and the associated cost of RBC genotyping in a large (>500 acute care beds) teaching hospital transfusing about 12,000 RBC/year.

Methods: All antibody investigations performed from Jan 1 2010 to June 30 2010 (6 months) at St-Michael’s Hospital were retrospectively reviewed against pre-determined criteria to evaluate whether genotyping would assist with investigation.

Results: Out of 9,578 antibody screens performed, 273 (3%) were positive. According to our criteria, 20/273 (7.3%) cases would benefit from genotyping. The patients’ characteristics were 60% female, median age 58.6 years. Genotyping would be useful to determine the predicted phenotype of a patient with warm autoimmune hemolytic anemia in 7 (35%), to determine the predicted phenotype of a recently transfused patient 5 (25%), to determine the predicted phenotype of a patient with a positive DAT 4 (20%), to determine the Rh status of a patient typed as RhD positive by serology and an apparent allo anti-D 1 (5%), to solve the discrepant RhD results from 2 different hospitals 1 (5%) and for miscellaneous other reasons 2 (10%). The cost of extensive genotyping for 20 cases/6 months is around $6000.

Conclusion: According to our criteria, integrating genotyping into blood bank policies in a teaching hospital is feasible with a cost of around $12,000 per year.

Keyword(s):
- RBC genotyping
Discarding Red Cell Units Because of Temperature Standards: A Survey of Ontario and Quebec Blood Banks

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Background: The primary reason for not returning issued red blood cells (RBC) units to inventory after 30 minutes (min) out of a temperature (T) controlled environment is to reduce the risk of bacterial contamination. There is no data supporting that such short periods outside of the required temperatures will cause excess bacterial growth. The purpose of this survey was to determine how blood banks (BBs) interpret the CSA standards.

Methods: A survey was emailed to all 151 BBs in Ontario and 89 BBs in Quebec asking questions about the interpretation of CSA standard 10.10.5 in their facilities in 2009.

Results: The response rates for Ontario and Quebec were 73% and 64%, respectively. In Ontario, 45% of BBs reported discarding RBC units returned more than 30 min after issue or at a T>10°C while the majority of Quebec BBs (65%) reported discarding RBC units when returned more than 30 min after issue regardless of the T. In Ontario, 71% of BBs used an instrument to measure the T of RBC units on return: 44% of these used an infrared thermometer. In Quebec, only 33% measured the T of RBC units on return: 61% of these used a validated thermometer. 33 BBs in Ontario and 29 in Quebec reported that at least 842 RBC units were discarded in 2009 due to out of range temperature requirements.

Conclusion: The interpretation of the CSA temperature standards varies amongst blood banks and many RBC units are discarded because of this standard. A practical study to determine if the temperature or time out of the temperature controlled environment used in these standards is valid could potentially result in a significant costs savings.

Keyword(s):
- RBC temperature
- CSA standard
- RBC discard
**Purpose:** The benefits to health care-associated facilities of adopting “green” business practices include reduced environmental impact, improved business sustainability, increased productivity, and operational savings. Here, we describe the ongoing process and impact of green practices implemented at Canadian Blood Services’ BC and Yukon Centre.

**Methods:** Since 2007, the Facilities Management team has implemented a range of environmental upgrades to the blood centre’s physical plant, based on an energy utilization audit. In 2010, with senior management support, a centre-wide “Green Team” was created, comprising staff from all key divisions, to help identify and implement other “green” initiatives. An initial in-house workshop was conducted by a David Suzuki Foundation Community Leader and the Suzuki at Work Program (www.davidsuzuki.org) was adopted. A centre-wide “Green Team” has identified other potential environmental processes to trim waste; further reduce energy needs; conserve water; optimize transportation; and create a healthier workplace.

**Results:** Between 2007-09, electricity, natural gas and water consumption by the blood centre was reduced by 10%, 33% and 27% respectively. Fluorescent tubes and paints are now recycled. Email correspondence has replaced fax transmissions for routine communications from the blood centre to hospitals, saving at least 30,000 paper sheets per year and the Facilities Management Work Order system is now exclusively through email. The blood centre now accepts return of all blood shipment box packing material (e.g. bubble wrap, packing paper, cardboard inserts) which is reused.

**Conclusions:** A facility-wide approach to identifying and implementing environment-friendly business practices has improved efficiency and saved money. Further savings for blood operators and hospitals are achievable by their broader adoption.

**Keyword(s):**
- green initiatives
- blood centre
- cost saving
- improved efficiency

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**Notes**
Cherie Mastronardi 1*, Sandra Weiss 1, Ken Scammell 1, Wai Ling Wan 1, Faith Hunter 1, Clare O’Reilly 1, Bonnie Monteith 1, Angela Poon 1, Emily Joyce 1, Scott Ackwood 1, Andrew Jones 1, Lynne Marquis-Boyle 1, Jennifer Joly 1, Cilla Perry 1, and Jason Acker 1

1 Canadian Blood Services

CBS netCAD Vancouver provides blood products from donors who have been deferred as well as testing support for transfusion science related research and strategically aligned CBS operations initiatives. A gap analysis identified key areas for improvement including: prioritizing requests for products and testing support, scheduling donors to meet demand, maximizing product distribution, defining CBS netCAD responsibilities, improving communication with stakeholders and donors, improving documentation for tracking purposes and implementing cost recovery. A multi-phased project was initiated in May of 2010. Internal and external stakeholders were identified and a communication plan was developed to engage stakeholders prior to implementing the new processes. A new procedure to prioritize and document requests using a scoring model, a new CBS web based document database [enterprise content management system (Documentum®)], and a new netCAD email address to facilitate communication between netCAD and its stakeholders was implemented in October 2010. A new scheduling tool will be implemented in February 2011, and a cost recovery process is slated for implementation by April 2011. The new streamlined processes implemented to date have resulted in improved efficiencies in the distribution of blood products to internal and external stakeholders for research purposes and will facilitate the utilization of the technical and scientific expertise of netCAD staff to improve the efficiency of CBS operations.

Keyword(s):
- transfusion science research
- process improvement
Purpose: A survey was developed by the Public Health Agency of Canada (PHAC) in 2010 to assess the impact of the Guideline for Investigation of Suspected Transfusion Transmitted Bacterial Contamination, published by PHAC in 2008.

Methods: A web-based survey was administered to the hospital blood bank staff to address their awareness of the Guideline, its usefulness and hospital practices used to investigate suspected bacterial infection cases. To compare pre- and post-Guideline practices, a set of questions was identical to those in a prior 2006 survey.

Results: Twenty-four (53%) respondents completed the survey: 92% indicated the Guideline had been useful and 63% reported their facility’s investigation protocol had been changed following publication of the Guideline. Compared to the 2006 survey, reductions in the proportion of hospitals using segments of bags after transfusion (51% to 21%) were noted. The proportion of hospitals performing a Gram stain also increased (53% to 79%).

Conclusions: The survey results indicated that the 2008 Guideline was useful for hospitals and resulted in changes that improved compliance with best practices.

Keyword(s):
- adverse transfusion reaction investigation
- transfusion-transmitted bacterial infection
- infection control – transfusion safety
- guideline
Assessment of Potential Screening Questions for Babesiosis Risk

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Canadian Blood Services

Babesiosis is a tick borne infection with increasing prevalence in the north eastern USA. It is not currently found in Canada, but could be in the future. Babesiosis is blood transmissible, and can be fatal. We have assessed the potential for screening questions to identify donors with travel risk or with risk from tick exposure in Canada (in the event that babesiosis ever became endemic in Canada). Screening questions could be used to identify donors for deferral or for screening should a licensed test become available.

**Methods:** An anonymous questionnaire was mailed to 40,000 donors in 2008 (20,000 first time and 20,000 repeat donors) with a 45.5% response rate. Donors were asked about travel to risk areas in the USA, and about participation in outdoor pursuits in Canada in which tick exposure was possible.

**Results:** Nearly half (49.1%) of donors had traveled to the USA in the last 12 months, and of these about one quarter (27.5%, 13.5% of all donors) had been to a state where there was some risk of babesiosis. Nearly half of donors have camped in the last year (43.5%), with 11% of all donors having camped in hike-in or boat-in campsites.

**Conclusion:** Deferral or testing of donors at risk of babesiosis due to travel to endemic areas in the USA or due to possible tick exposure in Canada (should babesiosis become a risk in Canada) would be problematic due to the large numbers of donors with these behaviours. In Canada only one case of transfusion transmitted babesiosis has been documented from a donor who had recently traveled to the US.

**Keyword(s):**
- babesiosis
- donor research
White Blood Cell Counts for Platelet Donors at Canadian Blood Services Based on Gender and ABO Subgroup

Canadian Blood Services Halifax NS

Background: Platelet donors at Canadian Blood Services (CBS) have their complete blood count (CBC) parameters obtained prior to each apheresis collection. This study will review data on the white blood cell (WBC) counts of platelet donors.

Method: Over a five year retrospective period, WBC counts were obtained from 285 donors who had not donated within 3 months of CBC testing. The Abbott Cell DYN 1700CS was used prior to July 2009 with the Sysmex-Poch 100i being used thereafter.

Results: Males accounted for 78% of the donors and females 22%. The WBC range (95% percentile limit) for all donors was 4.20-10.10×10⁹/L. All of the gender ABO subtypes were generally within this range with no statistically significant differences.

Conclusion: This data represents the range for WBC counts in a healthy cohort of platelet donors in Nova Scotia. The current range in use at CBS in this setting is 3.4-11.8×10⁹/L with local hospitals (Capital District Health Authority) using a range of 4.5-11.0×10⁹/L.

Keyword(s):
- white blood cell
- platelet donor
- ABO
- gender

Notes
Granulocyte Counts in Platelet Donors at Canadian Blood Services Based on Gender and ABO Subtype

Canadian Blood Services, Halifax NS.

Background: The complete blood count (CBC) for platelet donors is obtained prior to every platelet apheresis donation at Canadian Blood Services (CBS). This study compares granulocyte counts of 284 platelet donors by gender and ABO subgroup.

Method: Over a five year retrospective period, a CBC was obtained from platelet pheresis donors who had not donated within 3 months of testing. The Abbott Cell DYN 1700CS was used prior to July 2009 with the Sysmex-PocH 100i being used thereafter.

Results: Of the 284, 60 were female and 224 male. The granulocyte range for all donors was 2.30-7.20 x10⁹/L (95% percentile limits). Upper limits for granulocyte counts were lower in the following donors: type B (female 5.70 x10⁹/L; male 5.50 x10⁹/L), female type AB (6.00 x10⁹/L) and male type O (5.80 x10⁹/L).

Conclusion: This data represents the range for granulocyte counts in a healthy cohort of platelet donors in Nova Scotia. The current range in use for granulocyte count is 1.3-6.8 x10⁹/L at CBS and 2.0-7.5 x10⁹/L at local area hospitals (Capital District Health Authority).

Keyword(s):
- granulocyte
- platelet donor
- ABO
- gender
Assessment of Platelet Counts Based on Gender and ABO Subtype in Platelet Donors at Canadian Blood Services

Canadian Blood Services Halifax NS

Background: Canadian Blood Services (CBS) mandates that platelet donors have a complete blood count (CBC) performed prior to an apheresis platelet donation. This study will record the platelet counts of donors by ABO subtype.

Method: Over a five year period, a retrospective study was performed on 285 platelet donors who had not donated within 3 months of the time of CBC testing. The Abbott Cell DYN 1700CS was used prior to July 2009 with the Sysmex-PocH 100i being used thereafter.

Results: There were 224 male and 61 female donors. The range for all platelet donors was 177-339 X 10^9/L (95% percentile limits). Platelet counts for all subgroups of each gender were generally within this range.

Conclusion: This data is generally consistent with the range for platelet count used at area hospitals (Capital District Health Authority; 150-350 X10^9/L) but differs from the reference value for platelet donors currently used at CBS (150-500X10^9/L).

Keyword(s):
- platelet count
- platelet donor
- ABO
- gender
Hematocrit Levels in Platelet Donors at Canadian Blood Services Based on Gender and ABO Subtype

Canadian Blood Services Halifax NS

Background: Donors at Canadian Blood Services (CBS) must have a complete blood count (CBC) performed prior to each platelet apheresis donation. A hematocrit level of ≥ 38% must be present in order to be eligible to donate. This study will record the hematocrit levels of donors of both genders by ABO subtype.

Method: Retrospective five year study of 285 platelet donors who had not donated within 3 months of CBC testing. The Abbott Cell DYN 1700CS was used prior to July 2009; the Sysmex-PocH 100i was used thereafter.

Results: There were 224 male and 61 female platelet donors. The overall range for hematocrit was 0.37-0.48% (95% percentile limits). Female range was 0.36-0.43%; males 0.37-0.48%. Hematocrit values for all ABO gender subgroups were generally within the overall range.

Conclusion: This data demonstrates that the lower limit for hematocrit for both female and male platelet donors at CBS differs from the corresponding values currently in use at area hospitals (Capital District Health Authority; 0.42% for males; 0.37% for females).

Keyword(s):
- hematocrit
- platelet donor
- ABO
- gender
Determination Factors Defining Maximal Surgical Blood Ordering Schedule - A Recent Experience

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CLS/University of Calgary

Background & Purpose: Establishment of institutional specific maximal surgical blood order schedule (MSOBS) provides a guideline for an efficient use of available shelf life of any single unit of red cells. Monitoring crossmatch-to-transfusion (C/T) ratios is one of the main indices that has been used by MSOBS to meet the needs of 80 - 90% of surgical patients, however other indices such as transfusion index of <0.5 unit (<0.5 RC unit per surgical procedure) is also another suggested index for distinction of high risk patients. Other particular circumstances, including patient demographics must be also taken into the consideration. The amount blood held or dispensed to the operation rooms (ORs) usually impacts the optimized utilization of available blood. We decided to update the regional MSOBS based on the current surgical transfusion requirements.

Results: The regional annual data (2009-2010) for all surgical patients with dispensed blood were collected within four major Calgary metropolitan hospitals. Total number of 10434 units of red cells was dispensed to the ORs during the above period. From those only 2930 units of red cells had been transfused (28% of dispensed of red cells). The rest (7504 RCs; 72%) had been returned to the transfusion medicine. The percentage of surgical transfusion and Blood dispense to ACH, regional pediatric hospital were minimal (<2% of total regional transfusion and blood dispense). Therefore this hospital was excluded from MSOBS review. The percentage of returned or untransfused blood for two of three major hospitals were 65% to 78%, however one of the hospitals showed low rate of untransfused blood (24%). The hospitals with significantly higher percentage of returned blood were the major center for Trauma surgery/Cardiac surgery and vascular surgery respectively. From 27% of transfused patients only 10% of them had been transfused by 3 or more units of blood (Range; 3-18 units).

Conclusion: The study revealed that C/T ratio would be a good index to set a new MSOBS, however the transfusion index (% of transfused red cells <0.5) did not appear to be reliable in our patient population, however the percentage of transfused patients per specific surgical procedure (> 3) units was an reliable index for the distinction of high risk patients.

Keyword(s):
- MSOBS
- red cell utilization

Notes
Simulation of Blood Inventory Based on Red Cell Shelf-Life, Get Prepared for the Era of Fresh Blood Utilization

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CLS/University of Calgary

**Background & Purpose:** Prospective increase in the demand for fresh blood utilization has been the focus of recent studies showing high incidence of transfusion associated adverse reactions among patients transfused with aged red cells. Shelf-life of stocked red cells has been already reduced by some of national blood authorities. Limitations in supplement of fresh blood have raised serious concerns among blood suppliers and transfusion services. This study was designed to use a simulation model to understand the dynamics of blood inventory based on the red cell shelf-life and to find out about the capacity of blood system in providing fresh blood at high demand conditions.

**Method:** Recent historical and categorized red cell shelf-life at three levels of “regional CBS”, “regional hospitals” and “Transfused red cells” were used as a start point to simulate the blood inventory, changes in the distribution of red cell shelf-life as well as expiry rate at different levels of fresh blood utilization.

**Results:** The regional blood inventory was simulated at different levels of red cell stock inventory ranging from 7 to 23 days (Inventory index of 7 to 23 based on Red Cell Demand) at different levels of fresh blood utilization (<5%, 15%, 25% and 40%). The expiry rates at different conditions determined to define the inventory range and the maximum inventory without increase in the expiry rate.

**Conclusion:** Simulation model showed that red cell inventories regardless of inventory index/level reach a balance point within 8 to 10 weeks. After reaching a balance point, the distribution of red cell shelf-life stays at a relative fixed proportion. National and regional red cell inventory could hold red cell stock of up to 20 - 22 days without a significantly increased expiry rate, however to keep the expiry rate low, the major component of transfused red cells must be aged red cells within 5 - 6th weeks of shelf-life. With high national/regional inventory of >13 days, increase in fresh blood utilization to the amount of more 15% will be accompanied by increased expiry rate at inventories lower than threshold (Inventory index of 21). At optimal levels and well coordinated condition, the fresh blood utilization may be increased up to 40% without increased expiry rate.

**Keyword(s):**
- fresh blood utilization
- red cell shelf-life
- red cell inventory

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**Notes**
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**Background:** The Canadian Blood Services (CBS) Perinatal Testing Laboratory serves as a centralized service for prenatal serological testing of pregnant women in Alberta (AB), and North West Territories (NWT). Routine serological tests in pregnancy include ABO and Rh(D) blood typing as well as a screen for red cell alloantibodies and, if present, identification and titration of antibodies. Clinically significant antibodies are those that have been reported to cause HDN.

**Methods:** The number of antibody screens, total number of patients with antibodies, and antibody identification were determined for each year between 2004 and 2007 from published annual reports of the perinatal testing program. Live births and fetal deaths recorded in Alberta and NWT for the same period were determined from the Statistics Canada website. The number of patients with antibodies per 1000 births was calculated.

**Results:**

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<th>Year</th>
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<td>57040</td>
<td>60704</td>
<td>65482</td>
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<td>patients with antibodies</td>
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<td>277</td>
<td>234</td>
<td>248</td>
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<td>common CSA</td>
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<td>K,E,D</td>
<td>K,E,D</td>
<td>E,K,D</td>
</tr>
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<td>43137</td>
<td>46227</td>
<td>50120</td>
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<tr>
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<td>6.4/1000</td>
<td>5.1/1000</td>
<td>4.9/1000</td>
</tr>
</tbody>
</table>

**Conclusion:** Antibody prevalence amongst the prenatal population in AB and NWT is stable with similar frequencies of clinically significant antibodies over time.

**Keyword(s):**
- prenatal
- antibody prevalence

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**Notes**

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