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RÉSUMÉS
ABSTRACTS

PL-1

DENGUE VIRUS PERSISTS IN STORED PLATELETS AND RED BLOOD CELLS

Michael R. Sutherland[1], Ayo Y. Simon[1], Katherine Serrano[1], Peter Schubere[1], Jason Acker[2] and *Ed L. Pryzdial[1]

[1]CBS, Center for Innovation; Centre for Blood Research and Dept of Path and Lab Medicine, UBC, Vancouver, BC; [2]CBS, Network Centre for Applied Development, CBS, Vancouver, BC; Dept of Lab Medicine and Path, U of A, Edmonton, AB

Dengue virus (DENV), a positive-sense single-stranded RNA Flavivirus, infects ~390 million people annually. Significant Viral load is seen in asymptomatic individuals resulting in transfusion-transmission and a threat to blood systems globally. It is currently unknown whether DENV can maintain its viability or even replicate during storage of cellular blood products. Therefore, we investigated the persistence of DENV (serotypes 1-4) in stored platelet (PC) and red blood cell concentrates (RCC) under standard blood bank conditions. To mimic the maximal asymptomatic level, 105 infectious units/ml of purified DENV were spiked into donor-derived, leukoreduced PC or RCC produced at the Canadian Blood Services, Network Centre for Applied Development. Following inoculation, the PCs were stored for 7 days on a shaker at room temperature and sampled daily, while RCC were stored for 42 days at 1-6°C with weekly sampling. Infectious DENV was detected using cytolytic plaque assays immediately following addition to bath products. Viable DENV underwent logarithmic inactivation in PC by Day 1 post inoculation, however, between 1-5% of the initial infectious DENV was detected up to Day 7. The presence of DENV1-4 was confirmed by quantitative rtPCR, showing as much as a 7-fold increase in DENV RNA genome by Day 3, with continued detection throughout the 7 day storage period, consistent with platelets having known translational machinery to produce protein from mRNA like that of the DENV genome. Analysis of the RBCs by plaque assays showed viable DENV throughout the 42 day storage period, consistent with stabilization of the virus at the lower storage temperature. Interestingly, even though RBCs do not have the ability to translate mRNA, evidence of virus replication was observed. Platelets and leukocytes were below conventional detection methods, nevertheless together with residual reticulocytes may be involved. The possibility that DENV or other RNA viruses may persist and replicate in cellular blood products highlights the need to hasten the development and implementation of universal pathogen inactivation.

PL-2

USE OF PROTHROMBIN COMPLEX CONCENTRATES IN PATIENTS WITH HEPATIC COAGULOPATHY: A SINGLE CENTER RETROSPECTIVE STUDY

***Guillaume Richard-Carpentier[1], Normand Blais[2] and Benjamin Rioux-Massé[2]**

[1]Université de Montréal, QC; [2]Département d'hématologie et médecine transfusionnelle, Centre Hospitalier de l'Université de Montréal (CHUM) , QC

Background: The CHUM is a major reference center in hepatology and liver transplantation. Prothrombin complex concentrates (PCC) have been used in selected patients with liver disease in this center. This study aims to analyze the efficacy and security of this use.

Methods: A retrospective study was conducted by reviewing the medical records of all patients with liver disease who received PCC at the CHUM between January 1st 2009 and December 31st 2012. We collected the INR before and after the administration of PCC, searched for adverse events, and evaluated bleeding control.

Results: A total of 51 patients were included. Forty-one patients (80%) had cirrhosis and nine (18%) had acute liver failure. The status of liver disease could not be determined for one patient. Twenty-eight patients (55%) received PCC for bleeding and 20 patients (39%) received PCC before an invasive procedure. Adequate dose of PCC was used in 28 patients (55%). The INR was corrected to ≤1.8 in 30 patients (61%). Eight of ten patients (80%) corrected their INR in the Child-Pugh A group, compared to 10 of 13 (77%) in the Child-Pugh B group, 10 of 17 (59%) in the Child-Pugh C group and 2 of 9 (22%) in the acute liver failure group. Significantly less patients with Child-Pugh C cirrhosis and acute liver failure corrected their INR to ≤1.8 when compared to patients with Child-Pugh A and B cirrhosis (p=0.02; chi square test). Control of bleeding was achieved in 32% of patients (9/28) who received PCC for this indication. Three patients (6%) had thromboembolic events after receiving PCC. Four patients (8%) died within 24 hours of PCC administration but all the deaths were related to an underlying condition.

Conclusion: Our study showed that in patients with hepatic coagulopathy only a minority of bleeding events were controlled by the administration of PCC. Bleeding associated with hepatic coagulopathy is complex and the role of PCC requires further evaluation in regards to other blood products utilization and interventions.

SC-1

IMPACT OF OSTEOBLAST MATURATION ON THE EXPANSION OF UMBILICAL CORD BLOOD CELLS

Roya Pasha and Nicolas Pineault

Center of Innovation, Canadian Blood Service, Ottawa, ON

There is renewed interest in stem cell-based therapies following advances in expansion technologies and trials that demonstrated clear benefits such as accelerated neutrophil engraftment. Thrombocytopenia remains however an issue partly due to the lower dose of hematopoietic stem and progenitor cells (HSPC) during cord blood (CB) transplantation. We recently showed that osteoblasts derived from mesenchymal stromal cells (MSC), referred as M-OST, represent a superior source of feeder cells for the expansion of HSPCs and subsequent platelets engraftment in a transplantation model. We hypothesized that the capacity of M-OST to modulate the growth and differentiation of HSPC will vary as a function of their differentiation and maturation status. The objective of this study was to measure the expansion and differentiation of CB CD34+ cells cultured in conditioned media (CM) prepared with M-OST induced to undergo osteogenic differentiation for various length of time (3-21 days). Impacts on CB cell growth were assessed after 7 days of culture with the M-OST CMs by immunophenotypic analysis of the CB cultures. We also monitored the maturation of M-OST by FACS analysis, PCR and Alizarin red staining to characterize immunophenotypic changes during M-OST maturation. CB cell growth in M-OST CM generally increased as a function of the M-OST maturation, with the highest level of expansion obtained with day-21 M-OST CM (128±6, mean±SD, n=2) vs day-6 (79±5) vs standard control culture (17±1). M-OST maturation also significantly impacted the differentiation of the CB CD34+ cells in culture; there was a marked induction and/or promotion of myelomonocytic differentiation as a function of M-OST maturation (29±1%, 20±2% and 4±1% of CD14+ cells in day-21 CM, day-3 CM and control respectively) at the expanse of megakaryocytic differentiation. Phenotyping analysis of M-OST revealed stable expression of CD105, CD44, CD90 and CD166, and differences in CD63, CD73, CD146, OPN and ALP expression as a function of maturation. In conclusion, our results confirm that the differentiation and maturation status of M-OST have major influences on the modulatory activities of these cells on hematopoiesis.

SC-2

FIBRINOGEN BUT NOT FACTOR VIII LEVELS DETERMINE THE ABILITY OF TRANSFUSED PLASMA TO REDUCE BLEEDING IN A NOVEL MOUSE MODEL

Louise Eltringham-Smith, Xi Lei, Adili Reheman, Edward L. Pryzdial, Heyu Ni and *William P. Sheffield

Canadian Blood Services Centre for Innovation, Hamilton and Toronto, ON, and Vancouver, BC

Canadian regulators require transfusion services to determine coagulation factor VIII (FVIII) activity levels in transfusable plasma as a quality control measure. However, the relationship between FVIII activity and the effectiveness of transfused plasma at reducing bleeding is unclear. Purpose: To compare the ability of transfusion of plasma from wild-type (WT) , FVIII knockout (FVIII -/-), or fibrinogen knockout (fgn -/-) mice to reduce blood loss in coagulopathic mice. Methods: Anesthetized CD1 mice were rendered coagulopathic by four rounds of exchange of 0.5 ml whole blood for 0.5 ml washed red cells in 5% human albumin solution (HAS). Immediately prior to tail vein transection they received either 12 ml/kg HAS or WT, FVIII -/-, or fgn -/- mouse plasma. Shed blood was collected directly into warm water and quantified by spectrophotometry. Results: Compared to untreated mice, Blood Exchange-induced Coagulopathy Approach (BECA) mice exhibited reduced platelet counts (278 ± 130 vs. 825 ± 280 X 10 exp 9/L), reduced hemoglobin (78 ± 9 vs. 134 ± 8 g/L), and reduced coagulation factor activity compared to pre-BECA levels (e.g. 20 ± 6% fgn) (n=7 ± SD). BECA mice transfused with 5% HAS lost 270 ± 170 µL of blood, an amount reduced by either WT (70 ± 50 µL, $p<0.001$) or FVIII -/- (80 ± 60 µL, $p<0.01$) but not fgn -/- (210 ± 70 µL, $p>0.05$) plasma transfusion (n=15 ± SD). Mice not subjected to the BECA protocol lost 30 ± 30 µL of blood. Substituting plasma thawed and refrigerated for 5 days for freshly thawed plasma (FP) did not diminish its efficacy (compare blood losses of 80 ± 40 µl versus 80 ± 50 µl ($p>0.05$)). Conclusions: The content of fibrinogen, but not FVIII, was a relevant determinant of the efficacy of plasma transfusion in coagulopathic mice. Their residual FVIII levels appeared sufficient to support hemostasis without supplementation. Thawed stored plasma retained its efficacy, consistent with the greater stability of fibrinogen compared to FVIII. The regulatory focus on FVIII as a plasma quality marker should be reconsidered.

SC-3

ROLE OF CD62L IN THE INHIBITORY EFFECT OF IVIG ON THE CYTOTOXIC ACTIVITY OF CD8+ T CELLS

***Dominique Chabot, Patrick Trépanier and Renée Bazin**

Héma-Québec, Innovation R&D, and Laval University, Québec, QC

Background: IVIg is used for the treatment of an increasing number of autoimmune disorders including diseases in which self-reactive cytotoxic CD8+ T cells (CTL) play a major role. We recently showed that IVIg decrease the overall lytic activity of CTLs (Trépanier, Chabot et al, Immunology 2014). CD62L, an adhesion molecule expressed on naïve T cells, is cleaved following activation and its shedding from the surface of CTL is associated with the acquisition of both cytotoxic abilities and surface expression of the cytotoxic-associated marker CD107a. We thus postulated that IVIg modulates the expression of CD62L by CTLs to exert its inhibitory effect.

Method: C57BL/6 mice were injected twice with ovalbumin. Some animals also received multiple injections of IVIg (2.5g/kg). All mice were sacrificed after 28 days. Splenocytes were recovered and the expression of CD62L and CD107a on CD8+ gated T cells was evaluated using flow cytometry. The effect of IVIg on CD62L expression was also studied on human CD8+ T cells using PBMC incubated with or without IVIg (15 mg/ml) for 24 h.

Results: The expression level of CD62L on CD8+ T cells recovered from the spleen of IVIg-treated mice was approximately twice higher than that observed with untreated immunized mice. Conversely, the expression level of CD107a was approximately twice lower on CD8+ T cells from IVI g-treated mice compared to untreated mice. Similarly, our results revealed a statistically significant higher level of · CD62L expression on CD8+ T cells from PBMC of four different donors following incubation in the presence of IVIg.

Conclusion: IVIg treatment during the course of an active immune response is associated with a higher expression of CD62L and a concomitant lower CD107a expression. This suggests that the conversion of resting CD8+ T cells into fully effective cytotoxic T cells is impaired by IVIg. A similar effect of IVIg on CD62L expression by human CD8+ T cells was also observed. The control of CD62L may thus represent one of the mechanisms by which IVIg mediates its therapeutic effects in autoimmune disorders.

SC-4

ANTIBODY-MEDIATED IMMUNE SUPPRESSION INDUCED BY ANTI-RBC ANTIBODIES INCREASES IN EFFECTIVENESS WHEN MIXTURES OF MONOCLONAL ANTIBODIES ARE USED

***Lidice Bernardo Reyes[1], Alaa Amash[1] and Alan H. Lazarus[1,2,3]**

[1]Department of Laboratory Medicine at LKSK Institute of St. Michael's Hospital; [2]Canadian Blood Services; [3]Department of Medicine and Laboratory Medicine and Pathobiology, University of Toronto, ON

Background: The prophylaxis of hemolytic disease of the fetus and newborn (HDFN) is highly effective using plasma derived polyclonal anti-0. However, it would be beneficial to replace anli-D with a recombinant alternative. New therapeutics that comprise mixtures of monoclonal antibodies has been shown to be successful in the treatment of ITP. We have evaluated several monoclonal antibodies reactive with a model red cell antigen known as the HOD molecule in the induction of antibody rnediated immune suppression in a full murine model. HOD transgenic mice contain a RBC specific triple fusion recombinant protein comprised of hen egg lysozyme (HEL) in tandem sequence with ovalbumin and the complete human transmembrane Duffyb antigen that can be used to stimulate an immune response to the red cells.

Methods: HOD RBCs were transfused into C57Blf6 mice alone or together with selected combinations of anti-HEL antibodies (i.e., AMIS). Single monoclonal antibodies were compared to antibody mixtures binding to non-overlapping epitopes in their ability to induce AMIS. The resulting suppressive effect was assessed by evaluating the antibody response to the HEL protein by ELISA.

Results: Anti-HEL polyclonal antibodies completely suppressed the antibody response to the HOD antigen in C578L/6 mice while single monoclonal antibodies reactive with HEL were less suppressive despite using these monoclonal antibodies at high doses. Mixtures of monoclonal antibodies binding to non-overlapping sequences increased the AMIS effect in comparison to monoclonal antibody mixtures binding to overlapping sequences. Conclusion: The inhibitory efficacy of anti-RBC monoclonal antibodies could be improved by using antibody mixtures. The data show that by increasing the amount of antibody bound per RBC antigen, the greater the AMIS effect.

SC-5

FLOW CYTOMETRIC ASSESSMENT OF EFFICACY OF RH IMMUNE GLOBULIN (RHIG) PROPHYLAXIS AMONG WEAK AND PARTIAL D WOMEN

Julia Hendry, Joanne Luider, Iwona Auer and *M. Taher Shabani-Rad

Purpose: The rate of anti-D alloimmunization continues to be higher than expected despite widespread prophylaxis with RhIg. Noncompliant patients and presence of partial and variant weak D phenotypes have been assumed as possible aetiologies. This study was designed to investigate the interactions of RhIg with weak/partial D adult red cells and Rh-positive fetal red cells to provide evidence for or against the clinical indication of RhiG prophylaxis in these patients.

Methods: The study was performed using RC from weak D (5), partial D (2), Rh positive cord blood, and control RCs. Samples were exposed to diluted RhiG and analyzed by flow cytometry (FC) to measure the density of the D-antigen. Next, the specimens were incubated with RhIg washed and then mixed with Rh positive fetal 0 RCs at ratio of 1% and analyzed by (FC). Finally, the blood of weak/partial D patients was collected 3-4 weeks after receiving RhIg. DAT, PEG screen and FC were performed to test for the presence of bound and free RhIg on RCs and in plasma.

Results: Adult and fetal 0+ RCs expressed a Medium fluorescent intensity (MFI) of 40-50 due to bound RhIg. The MFI of Weak D and Partial D was 2.5-5.0 and 7-8 respectively. The mixtures of Rh positive fetal and weak/partial D maternal RCs showed a higher affinity of fetal red cells for bound RhIg. Moreover when fetal RCs were incubated with maternal RCs with no MFI, bound RhIg was detected on the fetal cells supporting the presence of low levels of maternal RC bound RhIg . Once released from the maternal cells RhIg was preferentially bound by fetal red cells. The specimens collected weeks after post RhIg administration showed the continued presence of bound RhIg on the maternal red cells and free RhIg in the plasma.

Conclusions: This experiment shows that Rh positive fetal RCs will compete favourably with weak/partial D maternal RCs by preferentially binding RhIg antibodies once equilibrium has been reached. In addition, this study supports the ongoing presence of RhIg on maternal RCs and in maternal plasma despite the RC binding capacity present in weak/partial D cases. The results support the use of RhIg prophylaxis in weak/partial D patients with unknown genotype.

SC-6

microRNAs IN BLOOD PRODUCTS: NEW QUALITY BIOMARKERS?

***Mélissa Girard, Marie-Pierre Cayer and Louis Thibault**

Héma-Québec, R&D, Québec City, QC

Purpose: MicroRNAs (miRNAs) are small noncoding RNA that negatively regulate gene expression or induce messager RNA degradation. Recent work has reported a diversified population of microRNAs (miRNAs) in red blood cells (RBCs) despite the absence of regulated gene expression in these enucleated cells. Modulation of miRNAs during storage of RBCs units has been observed but the fact that nonleukoreduced products were used does not allow asserting that these miRNAs originate from RBCs or from the contaminating leukocytes or platelets. Herein, we investigated the modulation of 6 miRNAs during storage of SAGM and AS-3 leukoreduced RBCs units.

Methods: Initially, RNA was extracted from AS-3 leukoreduced RBCs collected from 2 volunteers at day 1 and 42. RNA was reverse transcribed and subjected to microarray analysis. From these results, the most differentially expressed miRNAs, miR-16, miR-96, miR-103-2, miR-210, miR-720, and miR-1280, were selected. Levels of these miRNAs were measured by quantitative PCR (qPCR) in SAGM and AS-3 leukoreduced RBCs (n=6 per condition) on day 1, 7, 14, 21, 28, 35, and 42 days. Results were analyzed with the comparative Ct method using miR-491 as endogenous control for normalization.

Results: qPCR analysis revealed differential patterns for the six miRNAs during storage. miR-16, miR-96, miR-103-2, and miR-210 decreased during storage in AS-3 RBCs while their levels increase in SAGM RBCs. miR-1280, but especially miR-720, increase throughout storage interval, regardless of the RBC processing. A 6.8-fold increase was observed in SAGM RBCs between day 1 and day 42 whereas a 3.9-fold increase was observed in AS-3 RBCs.

Conclusions: These results suggest a modulation of miRNAs during storage of leukoreduced RBCs. Whole blood processing methods (e.g. solutions, temperature and storage time) could influence the levels of expressed miRNAs in blood products. Further work is however required to understand the implication of these miRNAs in maintaining the integrity and functionality of red blood cells. Nevertheless, this work opens new avenues towards identification of novel RBC quality biomarkers.

OP-1

EVALUATING THE IMPACT OF SHORTER SHELF LIFE FOR RED BLOOD CELLS WITH A GENERIC SIMULATION

***John Blake[1,2] and Matthew Hardy[1,2]**

[1]Dalhousie University; [2]Canadian Blood Services, Centre for Innovation

Background: Since the 1970's, red blood cells (RBC) have had a rated shelf life of 42 days. Recent transfusion medicine literature has raised questions regarding the safety of transfusing older red cells. Because of the impact on security of supply, a number of simulation studies have recently appeared in the literature evaluating the impact of a shorter shelf life for RBC. However, due to the different methods employed, it has been difficult to generalize the results of these studies.

Purpose: To identify, through the application of a generic simulation model, the impact of a shorter shelf life for RBC on a series of distribution networks.

Methods: A generic (or reusable) simulation framework was developed in Microsoft VB.Net that could be used to represent any regional distribution network in the CBS system. Specific instances of the model were the created to represent a small, medium, and large volume distribution network. Each model was validated and experiments were run in which the maximum shelf-life in each network was systematically lowered from 42 to 14 days.

Results: The impact of a shorter shelf-life for RBC was found to be dependent on the volume of materials handled by both suppliers and hospitals. Small sites and hospitals struggle at a 28-day shelf-life, while medium sized sites and hospitals can accommodate a 21-day shelf life. All sites, including large suppliers and hospitals, struggle at a 14-day shelf-life.

Conclusions: A shorter shelf-life will impact outdates, shortages, and the cost of operations in blood supply chains. The impact, however, is dependent on the size of the network involved. A universally acceptable shelf-life reduction cannot be identified.

OP-2

WHY NOT KEEP SOURCE PLASMA AT ROOM TEMPERATURE? A STABILITY STUDY AT 4°C, 22°C, OR 30°C

***Éric Ducas, Annie Jacques, Mélissa Girard, Claudia Bédard, Pierre Boisclair and Louis Thibault**

Héma-Québec, R & D, Québec City, QC

Purpose: Recently, Héma-Québec opened its first Plasmavie centre in Trois-Rivières (Qc) to meet the need of source plasma for the fractionation. In order to decrease logistical constraints and increase operation efficiency, the relevance of immediately keeping plasma units to 4°C until freezing after collection was questioned. Why not storing units at 20-24°C as is the case for the buffy coat method with whole blood? In this work, we studied the stability of important proteins in plasma stored at 4°C, 22°C, or 30°C for up to 72 hours.

Methods: Plasma was collected by apheresis. Each unit was split in three equal portions and stored for 72 hours at 4°C, 22°C, or 30°C. Aliquots were collected and frozen at -35°C. Levels of Factor VIII, fibrinogen, total proteins, albumin, and immunoglobulins (IgA, IgM, and IgG) were determined and compared.

Results: The 72-hour storage period before plasma freezing resulted in a marked decrease of the labile Factor VIII activity of 42%, 39%, and 45% for the plasma units stored at 4°C, 22°C, or 30°C, respectively. The storage length and temperature had no effect on levels of fibrinogen, total proteins, albumin and immunoglobulins (Table I).

Table I: Mean percentage of plasma protein activity following a 72-hour storage period at different temperature

	4°C	22°C	30°C
Fibrinogen (mg/dL)	93.9	94.9	90.2
Total Ig (g/L)	101.3	102.4	98.1
Total proteins (g/L)	97.8	98.6	99.5
Albumin (g/L)	99.0	99.5	99.7

Conclusions: Beside the expected loss of Factor VIII, the storage temperature of source plasma until freezing does not appear to influence the content of major plasma proteins important in the preparation of plasma fractionation products. Since its opening, Plasmavie center is storing source plasma units at room temperature.

OP-3

BLOODBRIEF: INCREASING AWARENESS AND INFLUENCING PRACTICE

***Cheryl Doncaster, Kathryn E. Webert, Virginia Gaffney and Lise Bourque**

Canadian Blood Services

Purpose: To promote optimal utilization of blood components/ products by heightening hospital awareness of issue trends over time.

Method: In September and December 2013, a BloodBrief was issued to top 50 hospital users of 0-negative RBC and AB plasma respectively. The BloodBrief was sent via e-mail to hospital transfusion committee chairs (or designate), contained three years of hospital specific issue data, ranking within the top 50 user list (and within peer groups) and anonymized issue data for other hospitals served by Canadian Blood Services. To determine effectiveness and impact, BloodBrief targeted hospitals were asked to complete an electronic survey in January 2014.

Results: A total of 64 hospitals were issued a BloodBrief (both 0-neg/AB plasma= 36, only 0-neg = 14, only AB plasma= 14). Follow-up survey respondents totaled 25 and represent 30 hospital sites. 60% of respondents, prior to receiving a BloodBrief, were not aware their hospital is a top 50 user of either 0-negative RBC or AB plasma. 68% indicated the BloodBrief highlighted data/information that was new (ranking within hospital peer group, top 50 user, 3-yr issue trends). The BloodBrief influenced 84% to review transfusion practice/ blood component demand with 48% of those anticipating revision to or development of transfusion policy (of these, 90% projected demand to decrease). Most hospitals opting to not review transfusion practice or policy cite issue data on par/below the national average and use of already very strict specific transfusion policy. Hospital suggested topics for future BloodBrief reports include: trends in trauma protocols, use of albumin, PCC, platelets, IVIG.

Conclusions: As a new approach to engage and inform hospital customers, the Canadian Blood Services BloodBrief is effective in promoting optimal utilization of blood components/ products. Heightened hospital awareness of issue trends over lime and comparisons within hospital peer groups influences hospital transfusion practice/policy.

OP-4

NATIONAL CONTINGENCY PLAN V ALIDATION

Nahrniak, S.[1], Lee, L.[1], Phua, C.W.[2], Mueller, D.[3], Morrison, D.[3], Callum, J.[2] anf Lin, Y.[2]

[1]University of Alberta; [2]University of Toronto and [3]Fraser Health Authority

Purpose: Blood contingency plans to deal with red cell shortages often recommend triage criteria and inventory management tools but validation is required to determine whether these recommendations will actually conserve units.

Methods: Alberta Health Services - Edmonton Zone (AHS-EZ), Royal Columbian Hospital (RC) and Sunnybrook Hospital (SB) validated the screening criteria in the National Plan and determined potential unit "savings" for all red cell requests between November 14th and 18th, 2013. The criteria included: bleeding status of patient, number of units requested, pre transfusion hemoglobin and elective surgical procedures.

Results: A total of 321 patients received 656 red cell units (RBC). AHS-EZ had 307 / 414 RBC (74%) issued with an initial Hb ≥70 g/L and 38 units for elective surgery (ES). Clinical review indicated only 171 units (41%) could be deferred. RC had 38 /68 units (56%) issued to patients with Hb ≥70 g/L and 7 units for ES but 20 RBC were for patients with bleeding so 26% of RBCs could be deferred. SB's major reasons for potential deferral included pre-transfusion Hb of ≥70g/L (33/41), multiple units requested inappropriately (6/41) and cancellation of elective surgery (2/41) accounting for 44% of RBC that could be deferred.

Conclusion: This exercise validated the basic criteria of hemoglobin cut off and bleeding status in the National Blood Contingency Plan. The percentage of units saved by deferral criteria were comparable between sites and overall was 35% of RBC units requested so would be useful in mitigating temporary shortages. Further evaluation would be required to accurately determine the safe duration of these deferrals. Although commonly cited as a conservation strategy, the cancellation of elective surgical cases would only have saved a total of 47 units (7% of total requests).

OP-5

SIMULATION AS A TOOL IN THE TRANSFUSION MEDICINE LAB TO EVALUATE AND IMPROVE THE MASSIVE HEMORRHAGE PROTOCOL

***Parrish, M.[1], VanSpronsen, A.[1], Shea, R.[2] and Clarke, G.[1,2]**

University of Alberta[1]; Alberta Health Services[2], Edmonton, AB

Background: Simulation is used as a highly effective educational technique to assess or instruct personnel in high stress and low opportunity situations over a wide variety of disciplines. Traditionally, simulation has not been used in training of laboratory staff within Alberta Health Services. The Massive Hemorrhage Protocol (MHP) is a process for delivering a large number of blood products to a critically bleeding patient within a short period of time. In hospital laboratories that see this type of patient infrequently, the MHP represents a high stress, low opportunity situation for laboratory staff. Our goal, therefore, was to use simulation as a means to assess the overall process of the MHP in the Edmonton zone allowing for practice, improvement and standardization across sites. In addition, we hoped to enhance the confidence and competence of technologists undertaking this task.

Methods: Several scenarios were created to mimic situations that technologists may encounter when an MHP is activated. Simulated blood products and patients were entered into the test system of the LIS (MiSys). Nurse educators and Pathologists were recruited to participate in their usual roles. Simulations were run with technologists at four hospital sites within the Edmonton area, each with varying experience. The total time from start until the MHP pack was ready to issue to a patient was recorded, as well as field observations of individual technologist actions. In addition to a verbal debrief, all participants filled out a written survey upon completion of the task.

Results and conclusions: There were several variations between sites and individual technologists. Based on observations and participant feedback, changes to the MHP procedure and training are planned, and are expected to improve the efficiency and standardization of the MHP process. The MHP activation is high stress and high stakes situation in a blood transfusion service. In order to increase the technologists understanding and to improve the effectiveness of the process within the laboratory we found simulation to be an ideal tool.

OP-6

EFFECTIVE USE OF AUTOMATED PANELS AND NOVEL WEB-BASED MIDDLEWARE TO SUPPORT CROSS-SITE INVESTIGATION OF RED CELL ANTIBODIES

***Mah, H., Chan, K., Danesin, P., Rizzetto, L., Roland, K., Sham, L., Tapp, S. and White, K. T.**

Chipperfield Transfusion Medicine Service, Vancouver, BC; Coastal Health, Vancouver, BC

Purpose: Many health regions have centralized antibody investigations at a hub site where there is additional medical and technical expertise in Transfusion Medicine. However, the sample travel time to the hub site can slow service delivery at the referring site. The purpose of this study was to evaluate whether a new workflow between a hub and referring site developed after the implementation of automation and web-based middleware would reduce the time required to complete antibody investigations.

Methods: NEO® and Echo® analyzers (Immucor) which use solid phase antibody identification panels (Capture-R® Ready-ID®) were implemented in November 2013 at two Vancouver Coastal Health hospitals. Web-based review of a single data repository from multiple instruments was made possible with the installation of ImmuLINKr»' (Immucor) middleware.The Lean Value-Stream tool was used to map a new workflow. One of the major opportunities involved moving the first panel testing task from the hub site to the referring site so that initial results are available sooner. Hub site technologists immediately review the panel results via ImmuLINKTM report and coordinate the investigation. Exclusions are performed with up to 2 additional automated panels tested at either the hub or referring site followed by manual techniques as required at the hub site. Mean turn-around-time (TAT) for high priority investigations was monitored for a 10 week period post implementation of the new workflow and compared to the mean TAT prier to the use of the automated systems.

Results: Mean TAT in the six months prior to automation was 4.6 hours. In the immediate post-implementation period, mean TAT dropped significantly to 2.1 hours. The correlation of TAT related to the complexity of the antibody investigation (e.g. single, multiple and pan-reactive antibodies) will be explored. Technologist satisfaction with the new workflow was high.

Conclusion: Technology enhancements can improve TAT for cross-site antibody investigations when a collaborative workflow is used.

CL-1

USE OF PROTHROMBIN COMPLEX CONCENTRATE (PCC) IN THE EMERGENCY DEPARTMENT: A QUALITY IMPROVEMENT STUDY OF ADMINISTRATION DELAYS

Marceau, D., Bordeleau, S., Poitras, J., Archambault, P. and Breton, C.

CSSS Alphonse Desjardins - HDL

Introduction: In some bleeding situations, quick reversal of warfarin anticoagulation is important, the delay can have an impact on mortality. This study aimed to improve the delay in administration when using PCC in the ED.

Methods: An audit and feedback quality improvement project was conducted in 3 phases: a retrospective phase, an analysis and feedback phase, and a prospective evaluation phase. The charts of all eligible patients in a ED who received PCC since November 2009 until October 2011 were retrospectively audited with pre-planned evaluation criteria.The administration delay of PCC was calculated from the time of prescription to the time of administration. After this retrospective chart audit, we determined where improvements could be attained, gave feedback to the ED and the blood bank, and created an action plan to ensure the timely administration of PCC.The action plan was then implemented in practice to reduce the administration delay. A 6 months prospective evaluation study was conducted to determine if our action plan was followed and if it improved the administration delays.

Results: 77 charts were reviewed in the retrospective chart audit. The mean administration delay was 73.6 min(STD [34.1]) with a median of 70min(25-75% IQR[45.0-95.0]). We found that the delays in a timely administration of PCCs were principally due to the following barrier: communication problems between the ED and the blood bank. We developped an action plan that involved the following elements: a flowchart to remind all clinicians how to order PCC and a new delevery method from the blood bank to the ED. During the 6 months following the implementation of our action plan, 39 patients received PCCs, and the mean administration time decreased to 33.2 min(STD [14.2]) ($p<.0001$) with a median of 30.0 min(25-75% IQ[24.3-38.8]).

Conclusion: This audit and feedback quality improvement project involving the implementation of an action plan reduced the administration time of PCC by more than half. We urge that similar studies be conducted to evaluate quality improvement strategies being newly.

CL-2

BABESIA AND HEPATITIS E SEROPREVALENCE IN CANADIAN BLOOD DONORS

Fearon, M.[1], *Scalia, V.[1], O'Brien, S.[1], Bigham, M.[1], Andonov, A.[2], Weger, S.[2], Bernier, F.[3], Dubuc, S.[3], Delage, G.[3] and Germain, M.[3]

[1]Canadian Blood Services Toronto, Ottawa, ON, and Vancouver, BC; [2]National Microbiology Laboratory, Winnipeg, MB; [3]Héma Québec, Montréal, QC

Purpose: Blood operators maintain vigilance for emerging infectious disease risks to the blood supply. In recent years, the protozoan parasite, Babesia microti has been recognised as a significant cause of transfusion transmitted disease in the northeastern U.S. Hepatitis E (HEV), commonly seen in the developing world, is now known to occur endemically in North America, and has been documented as a cause of post-transfusion hepatitis

Methods: The target was to recruit a total of 14,000 consenting blood donors (10,000 from Canadian Blood Services (CBS) from Manitoba, Ontario, and Atlantic Canada and 4,000 from Héma Québec (HQ). Babesia antibody (IgG) testing was performed by Indirect Fluorescence Antibody (IFA) assay at Imugen Laboratories, Norwood, MA. The National Microbiology Lab (NML) performed Hepatitis E Polymerase Chain Reaction (PCR) in pools of 50 on all donors. Hepatitis E antibody testing (IgG) was done on a subset of 4,000 donors using the Wantai HEV IgG ELISA. All donors filled out a questionnaire on possible risk factors.

Results:

CBS: All 10,062 CBS donors have tested Babesia antibody negative. Out of 3,071 donors tested for HEV and anti-HEV, there are 0 HEV PCR positives, and 110/2158 donors tested HEV IgG antibody positive (5.10% seroprevalence).

HQ: All 3924 HQ donors tested for Babesia antibody were negative and 131/1946 tested for anti-HEV were positive (6.63%); PCR tests results are pending.

Conclusions: Based on these results, exposure to Babesia microti seems to be rare in Canadian blood donors, at least in those provinces where they were tested. Hepatitis E antibody prevalence is consistent with that seen in other limited serosurveys in North America.

CL-3

CONSENT FOR BLOOD TRANSFUSION: DO PATIENTS UNDERSTAND THE RISKS AND BENEFITS?

Lani Lieberman[1,2,3], Daphne Cheung[2], Jeannie Callum[2,3] and Yulia Lin[2,3]

[1]University Health Network; [2]Sunnybrook Health Sciences Centre; [3]Department of Laboratory Medicine and Pathobiology, University of Toronto, ON

Purpose: Blood transfusion is a common medical intervention. The benefits, risks of and alternatives are often not consistently explained by physicians, nor understood by patients. Our hypothesis was that patients were not experiencing a standardized informed consent process, and that a consent video would assist in standardizing the information that patients receive and help improve patient understanding about blood transfusion.

Methods: Patients receiving their first transfusion at our centre were asked to complete a survey on their consent experience, then watch a standardized video on blood transfusion, and complete a follow-up survey. The standardized video was written and reviewed by transfusion medicine physicians.

Results: The survey revealed that the information recollected by patients from informed consent discussions was variable. Although 100% were given a reason why they were receiving transfusion, 85% recalled discussion about the benefits, 54% about potential reactions and only 24% were provided with information about alternative options. There were also variability in which potential reactions were discussed: 11 patients were told about severe allergie reaction, 9 about fever, 5 about heart failure and 3 about lung injury and bacterial contamination. Despite these findings, the majority of patients were comfortable consenting to blood transfusion. Patients felt that the video improved their understanding of the risks, benefits and alternatives to transfusion but did not change their comfort with consenting to blood transfusion.

Conclusions: Patients at our centre were not experiencing a uniform informed consent process prior to blood transfusion. Although the video improved their understanding of risks, it did not improve patient comfort towards giving consent for transfusion as the level of comfort was already high. The video is available available online at *http://www.youtube.com/watch?v=RxaPnLkgh-Oas* a resource for patients and physicians.

CL-4

TRENDS IN THE USE OF FEIBA AND RECOMBINANT FACTOR VIIA

***Kathryn Webert, Cheryl Doncaster, Robert Romans, Lise Bourque, Rachel Brace, Ahmed Coovadia, Jon Fawcett, Dorothy Harris, Valerie Paulson, Denyse Tremblay and Janet Unrau**

Canadian Blood Services

Background: Increasing amounts of recombinant factor VIIa (rFVIIa) and FEIBA were issued from Canadian Blood Services in Q1 and Q2 of fiscal year 2013/14. It was hypothesized that these agents were being stocked for bleeding associated with newer oral anticoagulants (NOAC).

Methods: A survey was sent to 387 hospitals across Canada (excluding Québec).

Results: 170 (44%) of hospitals responded. Only 24 hospitals (14.1 %) have a protocol in place for the reversal of Dabigatran. 9 protocols include the use of FEIBA, 14 include prothrombin complex concentrates (PCC) and 6 include the use rFVIIa. Other options included plasma (n=6) and vit K (n=2) or other (hemodialysis, tranexamic acid, transfusion of other blood products (red cells and platelets) as required, consultation with a specialist and transfer to another hospital). 14 hospitals (8.2%) have a protocol for the reversal of direct Xa inhibitors. 6 protocols include the use of PCC, 4 recommend FEIBA and 5 recommend rFVIIa. 4 protocols suggest plasma, 2 vitamin K, and 5 suggest other treatments (tranexamic acid, red cells and platelets, consultation with a specialist and transfer to another hospital). 34 hospitals (17.1%) stock FEIBA. 12 indicated thal FEIBA was a new addition to their hospital inventory (within 6 months). 33 hospitals indicated the amount kept in inventory which totaled 291,000 units or approximately 8819 units/hospital. 63 hospitals (31.7%) routinely stock rFVIIa. In 3 hospitals, rFVIIa had been added to hospital inventory within 6 months. These hospitals account for 1553 mg or approximately 25 mg/ hospital. Many respondents (n=102, 51.3%) stock neither FEIBA nor rFVIIa. 2 of these hospitals indicated that they planned to begin stocking either FEIBA or rFVIIa in the future.

Conclusions: The recent increases in issues of FEIBA and rFVIIa are unlikely to be due to hospitals newly stocking these products in case of bleeding associated with NOAC. Most hospitals do not stock FEIBA or rFVIIa so there is a potential for issues to increase in the future.

CL-5

EFFICACY AND SAFETY OF AMOTOSALEN-UVA PLASMA FOR TREATMENT OF ACQUIRED IMMUNE TTP

Herbrecht, R., Ojeda-Uribe, M., Kientz, D., Corash, L., *Cazenave, J.-P.

CHRU-Strasbourg, CH-Mulhouse, EFS, Alsace, France; Association ARMESA, Strasbourg, France; Cerus, Corporation, Concord, CA, USA

Therapeutic plasma exchange (TPE) for TTP requires large volumes of plasma (P) with risk of transfusion-transmitted infection (TTI). Pathogen inactivation (PI) of plasma with Amotosalen-UVA (INTERCEPT Blood System™ for Plasma (IBSP) may mitigate TTI risk. IBSP was implemented in Alsace, France in 9/2007. We compared the efficacy and safety of IBSP to Quarantine P (QP) for acquired immune TTP.

Methods: A retrospective, 2-period cohort study design was utilized. All patients with a clinical diagnosis of suspected TTP with pre-treatment ADAMTS13 assay data from 1/1998 to 3/2013 were reviewed, and patients with the following pre-treatment criteria were included: platelet count < 100 x 10e9/L, microangiopathic hemolytic anemia, serum creatinine< 354 1Jmoi/L, ADAMTS13 activity < 10%. Medical records were reviewed for data extraction. The primary efficacy outcome was the % of patients in remission (R) 30 days (d) after initiation of TPE. Secondary outcomes included the % in remission at d 60, d to remission, number of TPEs, volume of plasma to remission, and frequency (%) of relapse. Primary safety outcome was mortality at d 60. Secondary safety outcomes were: treatment emergent severe cardiac adverse events (AE) and serious AE (SAE). Results. 49 patients were screened and 31 qualified for review (Cohort 1 : QP=13; Cohort 2 : IBSP=18). Baseline demographics and disease severity between cohorts were comparable IBSP was comparable to QP, respectively, for efficacy outcomes: % R d 30 (61 v 46),% R d 60 (78 v 77), % relapse d 60 (7 v 40), TPE procedures (11.0 v 11.5), L/Kg of P to remission (0.47 v 0.39). However, median d to remission were significantly shorter for IBSP (15 v 24). Mortality was similar between cohorts (17 v 23%). Treatment emergent severe cardiac AE were not different (11 v 15%) nor were cardiac SAE (17 v 15%).

Conclusions: In this retrospective cohort study over 15 years, IBSP demonstrated comparable efficacy to QP for TPE of acquired auto-immune TTP. Severe cardiac AE and SAE were observed in both TTP cohorts, but all cause mortality was similar between cohorts.

CL-6

A REVIEW OF TRANSFUSION PRACTICES IN A RURAL HOSPITAL

Jacob Pendergrast and *Shadi Huladar

Background: Monitoring of physician transfusion practices can be performed through a variety of mechanisms, including both prospective and retrospective audit. To facilitate a retrospective audit at a small rural hospital, a physician pre-printed order form was designed to allow capture of relevant clinical information.

Methods: Pre-printed physician order forms for blood transfusion were developed by the Hospital Transfusion Committee and made mandatory. Descriptive data analysis was conducted after the fact using seven months of accumulated order sheets, with comparisons performed by two-tailed t-test. For the purposes of the current study, only red blood cell transfusion orders were analyzed.

Results: A total of 104 orders for red blood cell transfusion, written by 13 physicians for 53 patients was captured during the audit period. 30% of forms did not have a pre-transfusion hemoglobin documented, and 25% did not document the indication for transfusion. On average, 3.4 units were transfused per week at the hospital, usually with 2 units transfused at a time per patient. The average patient age was 72 and the infusion time per unit averaged 1.6 hours. 56% of patients received pre-medications, including 96% receiving diuretics. The pre-transfusion hemoglobin levels recorded by all physicians averaged at 77 g/L, but individual physician averages ranged from 65 to 94 g/L, indicating substantial variation in practice. "Signs and symptoms of anemia" was the most common indication for transfusion, followed distantly by treatment of active hemorrhage. Very few transfusion orders were written with the goal of preventing rather than treating symptoms. However, average pre-transfusion hemoglobin values were similar when comparing same-day and delayed transfusion orders, suggesting that most same-day transfusion orders were not urgent. Patients transfused for treatment of hemorrhage tended to have higher pre-transfusion hemoglobin levels than those with the stated indication of symptomatic anemia. It was also found that most RBC transfusion orders were written by a small group of physicians. However, there was no difference in average pre-transfusion hemoglobin level when comparing heavy versus light transfusers.

Conclusions: The use of pre-printed order forms can help identify inappropriate transfusion practices, but are limited by physician compliance in filling out the forms completely. Enforced completion of order forms may be necessary to make retrospective auditing a suitable alternative to prospective audit with approval.

1

WHAT IS THE TRUE COST OF A UNIT OF RED BLOOD CELLS? A COSTING MODEL FOR HOSPITAL USE

***Poseluzny, D., Lagerquist, O., Werstiuk, G., Ourdev, I., Slomp, J., Nahirniak, S., Clarke, G.**

NAIT and Alberta Health Services

Objective: The cost associated with delivering a unit of red blood cells (RBC) in Canada has been estimated based on costs related to collection, testing and delivery of blood products to hospitals. To our knowledge there has been no direct and thorough accounting of the cost of blood within a Canadian hospital in the past ten years. Determining current costs associated with providing blood transfusions requires in depth analysis of many complex activities within the hospital system.

Method: An activity-based costing model was developed to determine the cost of blood from the time of receipt at the hospital to the time of complete transfusion to a patient. Each of the steps involved in the transfusion process were mapped. The steps were sequentially monitored and timed on multiple occasions, and averaged. Materials, supplies, reagents, equipment used, and staff handling time were noted. Steps monitored included: inventory entry; storage; shipping preparation; shipping time; patient specimen collection; testing; issuing; delivery to a patient; transfusion time and; equipment and nursing time to monitor a patient. Final costs will be calculated based on the cost of labour, consumables and the relevant proportion of capital costs.

Results and conclusions: The activity-based costing model enables calculation of the hospital costs for RBC transfusion. The model is flexible in that each process step includes the resources and the associated costs; therefore steps can be added or removed depending on the particular processes in place at a facility. Future work will allow for validation of the model in hospitals of various sizes and with varying staff complements or process steps. Application of the model to other blood components and products will be assessed by future studies.

2

THE DEVELOPMENT OF PROVINCIAL CLINICAL AND TECHNICAL NEONATAL RECOMMENDATIONS

Nick Au[1], Susanna Darnel[2], Anita Gee[2] and Sheazin Premji[2]

[1]BC Children's & Women's Hospital; [2]BC Provincial Blood Coordinating Office

Purpose: To develop and provide provincial recommendations on Clinical and Technical Neonatal transfusion practice in British Columbia (BC).

Investigation: In 2011, the BC Transfusion Medicine Advisory Group (TMAG) proposed to form a provincial working group to review Neonatal Transfusion Practice in BC and develop new recommendations. Neonatal is defined as less than 4 months of age and includes areas of practice in the NICU and in non-NICU wards. One of the first steps of the review was to determine the current state in the province. The review and survey focused on the following:

1. Pretransfusion compatibility testing
2. Blood product selection and aliquoting
3. Emergency red cell transfusion
4. Blood product special requirements
5. Blood product administration

Six BC health authorities and the Yukon Territory participated in the survey and provided responses on common practices in their jurisdiction. Clinical and Technical sub-working groups were then formed. These two groups developed the recommendations based on findings from the provincial survey results along with completing a literature review.

Method: The BC Provincial Blood Coordinating Office (PBCO) facilitated the work of the Neonatal Transfusion Practice Working Group which included leaders from the BC TMAG, the BC Nursing Resource Group (NRG) and the Technical Resource group (TRG). The BC PBCO collected information, synthesized documents, and liaised between working group members.

Conclusion: Clinical and Technical Recommendations were simultaneously developed with cross representatives sitting on both sub working groups. Near final draft recommendations were circulated throughout the neonatal community in BC for feedback and comments. The final Clinical and Technical Recommendations for the Appropriate Use of Blood Components in Neonates and Infants Less Than 4 Months of Age were approved by the BC Transfusion Medicine Advisory Group for distribution and posting on the BC PBCO website in October 2013.

3

STAFF AND DONOR EXPECTATIONS BEFORE AND AFTER IMPLEMENTING EQUESTIONNAIRE

Osmond, L., O'Brien, S. F., and Goldman, M.

Background: eQuestionnaire was implemented in Ottawa and Sudbury in spring 2009. Pre- and post-implementation surveys assessed expectations and experience with the new system.

Methods: Clinic staff completed 53 pre- and 57 post-implementation anonymous paper questionnaires. Donors completed 160 pre- and 300 post-implementation face to face interviews.

Results:

Donors	Pre-imp. expectatIons	Post-imp. experience	p-value
Prefer screening with nurse	56%	32%	<0.0001
Easier to complete screening with nurse	33%	28%	0.5361
Faster process with eQue	58%	55%	0.7835
Satisfied with time it took to complete process on eQue		88%	

Staff	Pre-imp. expectatIons	Post-imp. experience	p-value
High training requirements	34%	80%	0.0014
More difficult for donors	47%	27%	0.0950
More time for other tasks	40%	9%	0.0001
Faster process for staff	32%	13%	0.0443

Conclusion: Donors don't perceive that eQuestionnaire made the screening process more efficient, but some did find it faster. They are satisfied with the system and at ease using it, and had fewer concerns once it was implemented. Clinic staff noted technical difficulties and higher than expected training requirements, without obvious improvement in efficiency. These findings suggest that implementation of eQuestionnaire would be eased by communicating realistic expectations, and by allocating resources for training and technical issues.

4

ASSESSING RESIDENTS' KNOWLEDGE IN TRANSFUSION MEDICINE IN FRENCH QUÉBEC UNIVERSITIES

Lavoie, M. and Boucher, M.

Hôtel-Dieu de Québec Hospital

Background: Physicians and residents of almost all specialties order transfusions every day without real formal training in transfusion medicine.

Methods: As a step to identify gaps in residents' knowledge in transfusion medicine, a questionnaire was emailed to all residents of the three French Québec Universities. The questionnaire had 20 questions about basic transfusion medicine knowledge and 7 demographic questions.

Results: 503 questionnaires were received and after exclusion of 110 incompletes questionnaires, the response rate was 14.4%. The mean score was 11/20 (range 4/20 to 18/20). The score improved with an increase of formal training hours, ranging from 8.6/20 with no formal training to 12.9/20 with more than 10 hours of formal training. The score also improved with each postgraduate year, ranging from 9.6/20 for PGY1 to 12/20 for PGY5 residents. The best score was by hemato-oncology residents with 14.6/20 and the worst score by pediatric residents with 9.1/20 although the questionnaire was aimed for knowledge in adult transfusion medicine. At the end of the questionnaire, 53.9% of residents believed that they definitely require more formal training in transfusion medicine compared to 26.5% at the beginning of the questionnaire and another 27% believed they probably need more formal training.

Conclusion: Marked knowledge deficits in basic transfusion medicine were noted. Additional formal training is required for a better utilization of blood product.

5

SAFE TRANSFUSION PRACTICE EDUCATION & COMPETENCY ASSESSMENT

Marilyn Collins, ART, Manager

Newfoundland and Labrador Provincial Blood Coordinating Program

Video (suitable for poster session or presenting the 35 min video session or Power Point presentation of the video and competency)

Purpose: To provide education to clinical nurses who facilitate transfusion of blood components. To provide competency assessment based on the education delivered in conjunction with prior basic knowledge. CSA Standards, CSTM Standards and Ontario Laboratory Accreditation (OLA) require competency, both initial and ongoing of all staff involved in the administration of blood and blood components.

Method: The video provides two scenarios depicting an allergie transfusion reaction and an acute hemolytic transfusion event. Throughout the scenarios, key elements associated with the recognition of signs and symptoms and reporting events are brought forward and elaborated upon. At the end of the video, a competency assessment is available in a pdf format or online by connecting to a website. The website is designed to have various levels of access for users, administrators and super-users.

Results: The user can complete the competency (3 attempts) and print a certificate upon successful completion. Failure to meet competency requires remedial discussion/instruction and one further attempt to meet competency upon approval by an administrator. Administrators are assigned by the super-users and can be updated regularly. The program is filtered according to Regional Health Authority, Site, Program. Also includes the date of completion of the competency which can be used annually to confirm renewal of required competency. Administrators can view the list and print ad hoc reports.

Conclusion: The educational experience will support continuying education and competency requirements required of staff involved in transfusion.

6

TRANSFUSION MASSIVE : SUIVI ET FORMATION CONTINUE

***Isabelle Beauregard et Chantal Robinson**

Chargées techniques de sécurité transfusionnelle, centre désigné des activités transfusionnelles de la Montérégie

But : S'assurer que les technologistes médicaux des 10 banques de sang de la Montérégie aient les connaissances nécessaires pour le bon déroulement d'un protocole de transfusion massive (PTM).

Méthode : Trois stratégies ont été mises de l'avant pour atteindre cet objectif. Premièrement, une présentation du protocole avec support visuel a été donnée, suivi d'un questionnaire d'évaluation des compétences. Deuxièmement, un compte-rendu a été mis en place et doit être complété par le technologiste dès la fin de chaque PTM. Finalement, afin d'assurer le maintien des connaissances, trois questionnaires ont été élaborés et distribués à tous les technologistes médicaux œuvrant dans les banques de sang.

Résultats : Ces trois stratégies permettent de s'assurer que les connaissances sont adéquates et de corriger rapidement toutes incompréhensions, que ce soit par exemple au niveau des notions théoriques ou dans le choix des produits sanguins.

Conclusion : En Montérégie, les PTM ne sont pas fréquents et les outils mis en place sont autant de moyens pour maintenir les connaissances à jour et ainsi assurer la sécurité des patients.

7

EVALUATION OF A TRANSFUSION EDUCATION RESOURCE FOR NURSES

***Gagliardi, K.[1], Evanovitch, D.E.[1], Sholapur, N.[2] Lane, S.[2], Young, L.[1], Lauzon, D.[1] and Thompson, T.[1]**

[1]Ontario Regional Blood Coordinating Network (ORBCoN); [2]McMaster Transfusion Research Program (MTRP)

Purpose: To develop an evaluation strategy that can be used to assess education resources, and to apply the strategy to an existing education resource that required revision.

Methods: An evaluation strategy was developed, using a recognized program evaluation framework. The framework identifies objectives, measures, information required, methods and sources. Both quantitative and qualitative methods were incorporated into the strategy. The evaluation framework was piloted on Bloody Easy Blood Administration (BEBA), an education program (in both paper and on-line formats), for nurses who transfuse blood. A 30 question survey designed in LimeSurvey was used to collect information from health care professionals who administer BEBA. In addition, qualitative feedback collected from 5419 participants who used the on line format was analyzed using the techniques of thematic analysis, facilitated by QSR International's NVivo 10 software.

Results: The survey, circulated to 160 health care professional contacts, included 60 hospitals not using BEBA. Fifty-five of the 100 hospital contacts using the program responded to the survey. 47% of the hospitals responding make the on-line program mandatory for new hires; 42% make it mandatory for existing staff. 43% reference the on-line program in orientation of nursing staff. 46% require that nurses complete the program annually. Thematic analysis of the participant evaluation responses resulted in the identification of 6 high level themes: subject matter; practice change; identification of learning needs; format; assessment; mandatory completion.

Conclusion: The evaluation report is in use by both the revision working group and the software company assisting with redesign. The survey provided evidence that the program is valued and should remain available. Issues identified by participants, with functionality and format, are proving helpful to a high quality revision process.

8

ASSISTING NURSING COMPETENCY IN BLOOD TRANSFUSION USING AN ELECTRONIC FORMAT

***Donna Touchie, Irene Oliveira and Antonio Giulivi**

The Ottawa Hospital

Purpose: Canadian blood transfusion regulatory standards require ongoing training for nursing staff who perform blood administration. We developed two electronic learning modules to assess blood transfusion competency of registered nurses at the Ottawa Hospital (TOH). We also evaluated one of these learning modules.

Methods: Two electronic learning modules were developed; Administration of Blood, and Blood Transfusion: Associated Risks. Key competency concepts were guided by TOH blood transfusion nursing policies and national blood transfusion regulatory standards. Two ten question quizzes were developed to test competency post completion of each module. The modules were launched on the TOH electronic teaching platform known as the Enterprise Learning Management System (ELM). A pilot study was developed to evaluate the content and utility of an electronic competency tool. Nurses working within the Deartment of Medicine were invited to complete the Blood Administration module and the associated ten question quiz. An electronic comment section was set up to capture qualitative feedback and assess completion times.

Results: Sixty three nurses participated in the pilot study within an allotted time frame. The majority of the comments were positive indicating the content was relevant, useful, and easy to follow. Many nurses indicated satisfaction with the electronic ELM format for ongoing learning and annual competency testing. The length of time for completion of the first module was collected. The completion times ranged from five minutes to forty minutes. A frequency distribution shows most nurses completed the module and quiz within ten to twenty minutes. The mean completion time was fifteen minutes.

Conclusion: Using the ELM system provides easy accessibility and convenience for TOH nurses to participate in ongoing learning about blood transfusion. ELM also allows managers to track staff learning. Annual competency assessments related to blood transfusion will reduce knowledge gaps related to safe practice, reduce errors, and meet regulatory standards for ongoing training.

9

LA FORMATION RÉGLEMENTAIRE CHEZ HÉMA-QUÉBEC : MEILLEURES PRATIQUES?

Beauséjour, A.[1] et Boutin, D.[2]

Direction de la formation réglementaire, Héma-Québec, Ville de Québec[1] et Montréal[2]

But : Le but de cette étude était de comparer la structure, le positionnement et la constitution de l'équipe de la formation réglementaire d'Héma-Québec avec les écrits récents sur la formation et d'autres entreprises qui attachent une grande importance aux normes de qualité et de sécurité.

Méthodologie: Une recension des écrits a été faite sur le rôle de la fonction formation à l'intérieur des organisations. De plus, un sondage portant sur les indicateurs prévus au grand livre de la formation a été envoyé à 14 entreprises pour faire cette étude comparative. Par la suite, une entrevue téléphonique semi-structurée a été accordée afin de clarifier les réponses données (n=6). Le même sondage a été complété par Héma-Québec. Les entreprises sélectionnées devaient avoir approximativement le même nombre d'employés, plusieurs sites de production et si possible une partie de leur personnel devait se déplacer à l'extérieur pour leur travail.

Résultats : Huit entreprises autres que Héma-Québec ont répondu au sondage (taux de réponse de 60 %). La majorité des entreprises avaient plus de 1000 employés pour lesquels des dossiers de formation devaient être gérés. Elles avaient également plus de 5 lignes de produits et plus de 3 sites de production. Dans la grande majorité des cas, le personnel de la formation relève de la direction des ressources humaines et les équipes sont constituées d'un peu moins de 10 personnes. De plus, 75 % des entreprises sondées avaient une structure hiérarchisée, ce qui est compatible avec le domaine réglementaire. La plupart d'entre elles ont des formateurs à temps partiel sur le terrain et ces derniers sont principalement choisis en fonction de leur capacité à transmettre l'information. Les formations spécifiques et techniques sont généralement développées à l'interne alors que celles reliées au relationnel et à la gestion sont bien souvent imparties. Malgré ce qui est recommandé dans les écrits, l'accompagnement post-formation est déficiente dans la plupart des entreprises balisées.

Conclusion : Des changements de structure ont été apportés à l'équipe de la formation réglementaire d'Héma-Québec pour être en phase avec les meilleures pratiques des entreprises similaires.

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DEVELOPING EDUCATIONAL RESOURCES TO ADVANCE ETHICAL UMBILICAL CORD BLOOD RESEARCH: A CANADIAN PERSPECTIVE

Carla Beak[1], Sophie Chargé[2], Rosario Isasi[1] and Bartha Maria Knoppers[1]

[1]Centre of Genomics and Policy, McGill University, Montréal, QC; [2]Centre for Innovation, Canadian Blood Services, Ottawa, ON

As the therapeutic use of cord blood stem cells in transplantation continues to grow, so too does the use of cord blood in research. From studies to improve cord blood collection, manufacturing and storing processes; to studies of the utility of cord blood to treat various hematopoietic and non-hematopoietic disorders; to the use of cord blood cells to derive pluripotent stem cells - cord blood research is making important contributions to the scientific and clinical advancement of the stem cell field. Obtaining an ample supply of such samples has been a challenge for the research community. In 2013 Canadian Blood Services (CBS) launched the National Public Cord Blood Bank (NPCBB) which collects, tests and stores cord blood units for use in transplantation. As part of their services, CBS is developing a system by which units that are not suitable for storage and transplantation are available to the scientific community for biomedical research purposes. To contribute to capacity building of Research Ethics Boards (REBs), who will be tasked with ensuring this research protects donors, we developed educational resources designed to assist REBs in the evaluation of research protocols which utilize cord blood samples. The "REB Primer on Research and Cord Blood Donation" (the Primer), outlines key ethical and legal considerations and identifies Canadian normative documents that are relevant to the use of cord blood in research. It also introduces CBS Cord Blood for research Program and describes the systems CBS is implementing to address governance issues. The Primer is intended to assist REBs in evaluating the ethical acceptability of research protocols, facilitate harmonized decision making by providing a common reference, and highlight the role of REBs in governance frameworks. However, it was written too to be accessible to the general public and may serve a broader purpose to increase public awareness of cord blood banking and the policies and procedures public systems have put in place to protect donors.

11

IMPROVING ACCESS TO TRANSFUSION MEDICINE RESOURCES THROUGH A USER FRIENDLY WEBSITE

***Sheazin Premji, Sonia Chau, Susanna Darnel, Jack Michaan and Vasco Vieira-Ribeiro**

British Columbia Provincial Blood Coordinating Office

Purpose: The British Columbia (BC) Provincial Blood Coordinating Office (PBCO) has a mandate to facilitate best transfusion practices in and to promote appropriate, safe, standardized and sustainable use of blood and blood products throughout BC. The PBCO works closely with transfusion medicine professionals, regional health authorities and Canadian Blood Services to fulfill its mandate. The PBCO website houses information that supports the transfusion care network in providing standardized best practice safe transfusion care, which in turn improves patient outcomes, improves blood utilization, minimizes inappropriate use, eliminates waste and improves service delivery. The website is a widely used tool by physicians, nurses, laboratory technologists and post-secondary schools to access information on transfusion medicine and practices including guidelines, forms, and educational materials. The current PBCO website was created in 2008; recent user feedback indicates that the website has opportunities for improvement, especially around content organization, navigation, and file and data sharing between advisory group members. The ability to share information and engage stakeholders is an essential part of the PBCO's work and an effective website is an important tool to enable this.

Method: Highlights of the new website include - updated and improved look and feel to enhance overall user experience; better navigation and search, including intuitive and relevant quick links for quick and easy access to frequently used documents and tools; reorganization of website content to better reflect the PBCO's current business lines and activities; improved usability with dedicated sections grouped by physician, technologist, and nurse; login section for members of stakeholder groups to enable quick and easy access to meeting documents and facilitate file sharing and access to data dashboards; section dedicated to education; mobile friendly version for smart phones and tablets to improve accessibility for clinicians on the go.

Results: The targeted launch of the new PBCO website is late Spring/early Summer 2014.

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MULTI-SITE IMPLEMENTATION OF NEW BLOOD BANK AUTOMATION AND WEB-BASED MIDDLEWARE APPLICATION (ImmuLINK)

***Sham, L., Chan, K., Chipperfield, K., Danesin, P., Mah, H., Rizzetto, L., Sheng, E., White, T. and Roland, K.**

Transfusion Medicine Services, Vancouver Coastal Health, Vancouver, BC

Background: In 2013, a plan was developed to implement new Blood Bank automation and a new novel web-based middleware application (ImmuLINK) in Vancouver Coastal Health (VCH). The new blood bank analyzers at three VCH hospitals would be integrated through the use of ImmuLINK which would consolidate test results for centralized review. Because this is the first time ImmuLINK would be interfaced with the laboratory information system (LIS), Sunquest Information Systems, many unique situations were faced in addition to ones expected from new analyzer implementation.

Purpose: To demonstrate the challenges that were faced in the implementation of new automation and middleware in a multi-site environment.

Discussion: The most difficult task during the implementation process was the coordination of technical personnel. This was facilitated by our Information Management Information Technology Services (IMITS) project manager who liaised with all personnel including staff from multiple sites, IMITS, Health Shared Services BC (HSSBC) hospital information privacy office, Sunquest, and Immucor (the vendor). Other technical challenges included: establishing communication between ImmuLINK, multiple blood bank analyzers, Immucor remote support, and the LIS; coordination with Sunquest in the development and testing of software drivers to accurately read information sent from ImmuLINK; creation and validation of test scripts to ensure that communication, operation, and quality assurances performed as expected; virtualization of the ImmuLINK server; and staff training at multiple sites on the new automation and software application. Two of the three VCH hospitals went live with the new automation and ImmuLINK in November 2013. To date, there are still on-going challenges to be overcome, including the implementation of new Blood Bank automation at the third VCH hospital.

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PHYSICIAN ORDER FORMS FOR TRANSFUSION MEDICINE- THE FIRST STEP TOWARDS COMPUTERIZED PHYSICIAN ORDER ENTRY

***Connie Colavecchia, Helen Downie, Lisa Merkley, Ana Lima, Yulia Lin and Jeannie Callum**

Sunnybrook Health Sciences Centre, Toronto, ON

Purpose: The Transfusion Error Surveillance System (TESS) indicates thal orders occur on the wrong patient and are incomplete, inappropriate (product and quantity) and lacking information required to ensure that special transfusion needs (e.g. irradiation, phenotypically matched) are provided to patients. To mitigate these errors and as a step towards computerized physician order entry (CPOE), Transfusion Pre-printed Order (PPO) forms were implemented.

Methods: Historically, nursing transcribed the physician's orders on to a manual requisition. Now the PPO must be completed by the physician and includes the diagnosis, indication, product type, quantity (dose) and transfusion rate as well as guidelines for ordering blood. A copy of the order is sent to the Blood Bank. The implementation of the PPO was piloted in the critical care units allowing for validation of the form and the new process in areas of high blood utilization. Prior to the implementation institution wide, training was provided to physicians and clinical educators.

Results: Data available for the first three months of using the PPO reflects the learning curve associated with a new process however preliminary data indicates no increase in inappropriate transfusion orders. When completed, the PPO provides information regarding indications and special needs to ensure patient receives the appropriate product in the appropriate dose.

Conclusions: The PPO is an example of a proactive measure of mitigating inappropriate transfusion orders however, until CPOE with physician assisted algorithms can be introduced, it will be difficult to see an improvement in appropriate transfusion orders. The PPO has validated a template on which the CPOE can be designed to assist clinicians and improve patient safety.

14

EVIDENCE OF TRULY INTERPROFESSIONAL COLLABORATION

Shanta Rohse

Canadian Blood Services

Purpose: Interprofessional collaboration (IPC) has become the standard of healthcare around the world. Yet, we lack a deep understanding of what it means for collaborative practice to be truly interprofessional and what factors nurture it. This study examined the experiences of a group of MLTs who participated in the 2011 and 2012 BloodTechNet Learning Competitions. The competition is a professional development program offered by Canadian Blood Services to foster IPC skills in transfusion services across Canada.

Methods: A series of online discussions among applicants and participants during the competitions were analyzed for themes involving IPC. Project plans, also stored online, were studied (1) to provide context for some of the discussions, such as timelines, resource needs and milestones; (2) to identify evidence of emerging collaborations; and (3) to triangulate with data collected from the discussions boards, such as whether or not the applicants acted on any potential collaborations that emerged during the discussions. Data were categorized, aggregated and interpreted following Creswell's method for patterns related to two research questions: (1) Was there evidence of truly IPC? and (2) What factors facilitated or inhibited collaboration?

Results: The online text discussions, viewed through the lens of interprofessional literature, revealed only one instance of truly IPC. In this instance, the applicant made fundamental changes to their project plan in response to feedback in the discussions. The result truly exceeded the "sum of its parts." Other projects were revised, some many times, but there was no evidence of truly IPC. Factors supporting IPC included interest, openness and flexibility among participants. Factors inhibiting IPC were too much team diversity, planning logistics and organizational support.

Conclusions: Truly IPC demands significant effort and learning from its participants. It is not surprising that the evidence revealed only one instance of truly IPC among the submissions. Organizational support was the most important factor. Senior management commitment is essential if teams are to benefit from IPC. These findings are part of a larger study that views IPC as relationships between individuals and larger social systems.

15

USE OF A REMOTE ALLOCATION BLOOD FRIDGE WITHIN A LARGE TRANSFUSION LABORATORY

***Megan MacQuarrie, Janice Hawes, Stephen Fitzgibbon and Christine Cserti-Gazdewich**

Purpose: The BloodSafe® Hemonine® Refrigerator (Haemonetics) is a remote blood allocation device functionally integrated with transfusion laboratory (BTL) processes. We aim to demonstrate merits of repurposing this device for internal use within our BTL at Toronto General Hospital (~1450 RBC/month by electronic crossmatch (EXM)).

Methods: Conventional BTL blood issue sequence (CBIS) vs internalized remote issue device (IRID) workflow metrics were compared.

Results: Prior to IRID, EXM was restricted to medical laboratory technologists (MLT). After IRID, all BTL staff (including technicians [LT]) could perform EXM. This associated with greater LT satisfaction & productivity, at 27% of attributable EXM, while freeing MLTs for advanced tasks. With the touch screen process, EXM was at least 2.5X faster at 53s. Time on IRID maintenance (30 min/day) did not exceed that which is gained from its efficiency but will expand with planned implementation of 2 more devices. As IRID does not capture product destination, applicable utilization metrics may be challenged.

Conclusions: IRID offered major advantages, enabling greater staff engagement, speed, and competency in EXM activities.

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EMERGENCY FRAMEWORK SIMULATION EXERCISE

Nahirniak, S.[1], Lee, L.[1], Phua, C.W.[2], Mueller, D.[3], Morrison, D.[3], Callum, J.[2] and Lin, Y.[2]

[1]University of Alberta; [2]University of Toronto; [3]Fraser Health Authority

Purpose: The National Advisory Committee created an Emergency Framework For Rationing Of Blood Products In Massively Bleeding Patients In A Red Phase Shortage (the Framework) using evidence based predictors of massive hemorrhage and mortality in a variety of clinical setting but the applicability of the triage criteria and potential unit savings of these criteria required validation.

Methods: In November of 2013, Alberta Health Services-Edmonton Zone (AHS-EZ), Royal Columbian Hospital (RC) and Sunnybrook Hospital (SB) performed an exercise to validate the Framework. Any patient in whom massive hemorrhage was identified over the study period were further evaluated for triage stopping criteria fulfillment, number of units transfused and survival outcomes.

Results: During the exercise, six patients were evaluated but only one MH patient met the triage stopping criteria.

Patient	Indication	Triage criteria met?	Number of units	Survival outcome
1	Ruptured AAA	No	8	No (<24h)
2	Trauma-MVC	Yes	3	No (day 12)
3	Post of bleed	No	21	Yes
4	Sickle crisis	No	6	Yes
5	Colon perforated	No	34	Yes
6	Ruptured AAA	No	29	No (<24h)

Discussion: Unfortunately, the period of evaluation was insufficient to capture a broad spectrum of massive hemorrhage patients to allow accurate validation and estimations of unit savings of the criteria in the Emergency Framework. Additional simulation exercises of longer duration are required for accurate validation but possible improvement in rupture AAA criteria may be required.

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INTRODUCING BARRIERS TO PREVENT ABO INCOMPATIBLE TRANSFUSION

***Helen Downie, Connie Colavecchia, Lisa Merkley, Ana Lima, Yulia Lin and Jeannie Callum**

Sunnybrook Health Sciences Centre

Purpose: Analysis of data obtained through error reporting systems identify that a high number of sample collection (SC) errors occur that have the potential to result in an A80-incompatible transfusion. The purpose of this report is to describe the implementation of process improvement initiatives to prevent ASO incompatible transfusion and ultimately improve transfusion safety.

Methods: Error tracking and data collection were performed using the Transfusion Error Surveillance System (TESS). Process improvement initiatives included pre-printed patient 10 labels in the patient's chart for sample labeling trialed in the Emergency department (2000), collection of a second independent sample as confirmation of blood group (November 2005), use of a specific tube for the confirmatory sample distributed only from the Blood Bank and only when required (January 2011).

Results: Over 2005-2012, there was a total of 6371 sample collection errors. Total SC related errors increased steadily from 1:61 in 2005 to 1:21 in 2012. This was likely in part due to introduction of process changes which increased detection of errors as well as changes in definitions of what constituted an error, However, there was a total of 979 high severity errors which also increased over time from 1:694 in 2005 to 1:147 in 2012. Despite this, no ABO-incompatible transfusions were reported.

Conclusions: The rate of high severity sample collection errors had significantly increased over time. However, no ABO incompatible transfusions were reported. Although our process improvement initiatives did not decrease errors, we believe that the initiatives increased the detection of these errors and thus created barriers to potential ASO incompatible transfusion. Our ultimate goal is to have positive patient ID technology being used throughout our institution thus eliminating the need for the confirmatory sample.

18

STATISTIQUES ET OPTIMISATION DE L'INVENTAIRE DES CULOTS GLOBULAIRES

***Chantal Robinson et Isabelle Beauregard**

Chargées techniques de sécurité transfusionnelle, centre désigné des activités transfusionnelles de la Montérégie

But : Optimiser la gestion de l'inventaire des culots globulaires en limitant la péremption et en favorisant la transfusion isogroupe pour les 10 centres hospitaliers de la Montérégie.

Méthode : Dans un premier temps, un outil statistique a été élaboré principalement pour faire ressortir les déséquilibres entre le groupe sanguin des receveurs et le groupe sanguin des culots globulaires transfusés. Ces statistiques sont extraites annuellement du logiciel provincial Trace Line. Dans un deuxième temps, chaque banque de sang réajuste ses seuils d'inventaire en fonction de ces statistiques tout en tenant compte de leur réalité.

Résultats : Ce rapport annuel permet aux responsables des banques de sang de constater l'utilisation qu'ils font des culots globulaires par rapport à la population desservie. Ce constat permet de réajuster plus efficacement les seuils d'inventaire et de mettre en place des stratégies pour atteindre les objectifs fixés telles que l'implantation du transport inter-hospitalier et la sensibilisation des technologistes au processus de gestion d'inventaire. Tous ces ajustements ont un impact régional que l'on constate par l'amélioration de plusieurs indicateurs de gestion tels que les taux de transfusion isogroupe et de péremption.

Conclusion : Cet exercice annuel permet l'ajustement et l'optimisation de la gestion de l'inventaire locale et régionale tout en offrant aux receveurs de la Montérégie le meilleur produit sanguin possible.

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RED BLOOD CELLS (RBC) AUDIT AT FIVE COMMUNITY HOSPITALS

***Lin, Y., Collins, A., Elahie, A., Evanovitch, D., Harrison, L., Saidenberg, E., Lafond, G., Lauzon, D., Thompson, T. and Owens, W.**

The Ontario Regional Blood Coordinating Network (ORBCoN) is a provincial organization dedicated to promoting best practices in transfusion medicine. ORBCoN has recognized that physician ordering practice is an essential area of focus for ensuring appropriate blood utilization, with RBC transfusion being the predominant transfusion activity of any hospital transfusion service. As a first step to understanding the educational needs of physicians, a baseline audit of RBC transfusion practice was conducted. An electronic audit tool was developed to capture information related to: ordering physician specialties; location of the transfused patient; and physician ordering practices. The audit was performed at five community hospitals in Ontario and captured 455 consecutive RBC transfusion orders during two seven-day periods. 856 RBC units were transfused to 384 patients. 55% of transfusion orders were two unit transfusions. 32% of orders were associated with a pre-transfusion hemoglobin 80 g/L or above, and 29% of posttransfusion hemoglobin values were 100g/L or above. The top prescribers were Internal Medicine, Emergency and Family Medicine physicians accounting for 54% of RBC units transfused. The results of the audit will be used to provide focused education for physicians in community hospitals.

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TRENDS IN THE ISSUES OF BLOOD PRODUCTS FROM CANADIAN BLOOD SERVICES

***Kathryn Webert, Dan Shavrnoch and David Howe**

Canadian Blood Services

Methods: Canadian Blood Services issues data for transfusable components for the past 10 years were examined to determine trends in product issues that reflect utilization. Third quarter (Q3) fiscal year (FY) 2013/14 data are estimates of full year FY2013/14 results.

Results: The number of number of red cell (RBC) units issued per 1000 population grew from 2006/7-2008/9 but has decreased over the past 6 years from 32.8 in FY 2008/2009 to 29.9 in Q3 FY2013/14. This equates to an absolute decrease of 28,065 units. 0-negative RBC issues have increased consistently over the past 10 years and now represent 11.9% of RBC issues. Platelet issues have begun to decrease since 2011/12 and in the most recent FY, 4.2 units were issued per 1000 population. This represents a decrease of 4,912. Platelet units requested cytomegalovirus (CMV)-negative have consistently decreased since 2007/8 from 36.7% of issues down to 29.5% in Q3 FY 2013/14. Plasma equivalent unit (1 apheresis unit or 2 frozen plasma units) issues have decreased consistently since 2006/7; from 10.11 units per 1,000 population down to 6.29 units in Q3 FY 2013/14. This represents an absolute decrease of 82,191 units. Group AB plasma has increased in issue with 8.93% of plasma issues being AB in 2006/7 and 11.31% in Q3 FY 2013/14. Issues of cryoprecipitate have increased since 2007/8 with 1.64 units per 1,000 population issued in FY 2007/8 and 2.26 units in Q3 FY 2013/14. In absolute numbers, this represents an increase of 19,601 units.

Conclusions: Review of Canadian Blood Services issue data reveal the following trends: decrease in RBC utilization with relative increase use in 0-negative RBC; decrease in platelet utilization as well as CMV-negative platelet utilization; decrease in plasma utilization with relative increase in group AB plasma use; and increase in cryoprecipitate utilization.

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A "TRANSPARENT BLOOD INVENTORY" REPORTING SYSTEM TO BETTER MANAGE RED BLOOD CELL INVENTORY DURING A BLOOD SUPPLY DISRUPTION

Janet Unrau, Judy Hrytzak and Mark Bigham

Canadian Blood Services, BC; Yukon Region, Vancouver, BC

Purpose: Since 2010, as part of the British Columbia (BC) Blood Contingency Plan, Canadian Blood Services (CBS), the BC Provincial Blood Coordinating Office and Health Authorities in BC have collaborated in a provincial transparent blood inventory (TBI) system. The benefit of a TBI was reaffirmed during a recent, planned disruption in routine blood supply to hospitals.

Methods: The TBI is a web-based blood inventory reporting system for RBC and platelet inventory held at CBS and at each of 21 hospitals across BC and Yukon, which account for a cumulative 83% of red blood cell (RBC) and 98% of platelet issues from CBS BC and Yukon Centre. In preparation for a planned 2 day disruption in routine CBS operations (16-17 Feb 2014), CBS staff worked closely with hospitals to ensure that hospital RBC inventories were at optimal levels to meet expected demand during and after the disruption. Participating institutions reported RBC inventory to the TBI just prior to the planned blood supply disruption.

Results: On 14 Feb, the TBI indicated that RBC inventory levels were above optimal inventory target at all 21 hospital sites, with these hospitals cumulatively holding 33% more RBC inventory than at CBS, and 57% of total shared RBC inventory. The TBI also revealed overall provincial RBC inventory just prior to the planned CBS service disruption at 37% above routine provincial target. Thus, although CBS RBC inventory level was below optimal inventory target, the TBI provided assurance that adequate RBC inventory was held at hospitals to meet anticipated demand during and following the disruption. Only 21 hospital RBC orders required manual issue from CBS BC and Yukon Centre during the service disruption.

Conclusion: TBI was a useful adjunct tool, providing assurance of adequate provincial RBC inventory during a short term supply disruption.

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ADJUSTED 0 NEG UTILIZATION RATIO (AOUR), A NEW INDICATOR FOR DEFINING OPTIMAL 0 NEG INVENTORY LEVELS

***Julia Hendry, Colleen Young, Joanna McCarthy and M. Taher Shabani-Rad**

Purpose: Due to the use of 0 neg red cells (RCs) as unmatched blood, shortages for 0 neg RC occur frequently. This study was performed to review the efficacy of current indicators in monitoring and management of 0 neg RC inventory and to develop a reliable indicator which may be used as a monitoring tool at different sized health care facilities.

Methods: Two commonly used indicators; 0 neg RCs demand/1 000 population (indicator A) and 0 neg RC Utilization/Total RC Utilization% (indicator B) were considered by this study. The practicality of these indicators to determine appropriate 0 neg RC inventory was assessed based on the historical red cell demand and size of health care facilities. Determination of appropriate use of 0 neg RC was not in the scope of this study. In addition a new indicator AOUR was developed and the accuracy and practicality of this indicator was compared to the conventional indicators.

Results: Indicator A was applicable and accurate at the Provincial level (11.5%) with no significant difference from the average national percentage (11.62%). The application of this indicator at individual hospitals was not practical due to population overlap. Application of indicator B to hospitals was not helpful to separate centres with adequate 0 neg inventory from those with inappropriate levels. This indicator fails for medium and small facilities due to the variability in RC demand and minimum inventory requirements. AOUR in combination with size classification of health care facilities was able to detect all facilities with inappropriate 0 neg inventory. Analysis showed that an AOUR of <0.2 is the appropriate ratio for centralized transfusion services. The appropriate ratio for large, medium and small hospitals are <0.5, <1.0 and <5.0 respectively. AOUR is not applicable to small health care centres with minimal RC demand which account for <5% of national RC demand.

Conclusion: The current 0 neg monitoring indicators are helpful at the national and provincial levels but they cannot be used to define the optimal 0 neg inventories at individual hospitals. The new indicator, AOUR is a reliable indicator for defining appropriate 0 neg inventory for most facilities.

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0 NEGATIVE RED BLOOD CELL TRANFUSIONS TO NON-O NEGATIVE PATIENTS

***Melanie Tokessy and Doris Neurath**

Eastern Ontario Regional Laboratory Association, The Ottawa Hospital

Purpose: In Canada, inventory of 0 negative red blood cells (RBC) frequently falls to critically low levels. Monitoring 0 negative RBC utilization has the potential to decrease inappropriate use through stricter policy implementation and education of laboratory staff and physicians. Data from 2013 was analyzed to gain understanding of 0 negative RBC utilization at our large tertiary care institution.

Method: 0 negative RBC transfusion data was retrieved from the laboratory Information system and categorized by reasons for transfusing to non-O negative recipients.

Results: In 2013, 2060 0 negative RBC were received into inventory; of those 1809 units were received from the CBS and 251 units were transferred through redistribution from our smaller site hospitals. A total of 1013 (49%) 0 negative RBC were transfused to non-O negative patients: 444 (43.8%) units were to sickle cells patients; 211 (20.8%) to patients with one or more antibodies; 165 (16.3%) were used to prevent unit outdating; 92 (9.1%) to BMT patients and 58 (5.7%) to neonatal patients. Only 21 (2.1 %) units were transfused as unmatched blood and 22 (2.2%) for other reasons.

Conclusion: This audit showed that almost 50% of 0 negative RBC were transfused to non-O negative patients. We recognize that strategy development is needed to ensure 0 negative RBC are appropriately reserved for those patients where no alternative is possible. Radical change in the current practices must occur in order to reduce and possibly eliminate the use of 0 negative RBC for non-O negative patients.

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GROUP 0 RBC UTILIZATION IN NON-GROUP 0 PATIENTS G

Clarke, G., Blain, H., Onell, R. and Nahirniak, S.

Alberta Health Services-Edmonton Zone

Purpose: Standards require use of group 0 red cells for computer crossmatch unless two blood groups are available to confirm the ABO typing of recipients. Historically, we performed two independent tests on the same specimen prior to providing group specific products. Following a near miss event, the policy was changed to require two independent specimens tested prior to providing group specific units (Confirmatory ABO or CABO). This raised the concem of increased group 0 utilization especially in emergency and massive hemorrhage patients without a historical blood group.

Methods: Group 0 red cell utilization was captured on month end reports for 6 months pre and for 11 months post protocol change which occurred in February of 2013. The proportion of group 0 units over total units was calculated. For each patient receiving group 0 units who was not blood group 0 (OTNO), their case was reviewed to determine if the reason was for CABO; antibody management; inventory management; unmatched, neonatal transfusion or other reason.

Results: Prior to CABO implementation, 40% of OTNO units were issued to emergency/trauma patients as unmatched; 35% congenital cardiac neonatal patients; 20% multiple antibody patients and 5% for hemoglobinopathy (reflecting CBS antigen typing availability). Although the total number of red cell units transfused overall has been consistent, there has been a very slight increase of OTNO. Prior to the new CABO process, 2.97% of our total red cells transfused were to nonO patients compared to 4.47% afterwards. The post CABO indication breakdowns have changed minimally but are continuing to be monitored.

Conclusion: Although there is a slight increase in group 0 utilization, the impact of confirmatory ABO testing is minimal. There continue to be many other reasons for transfusion of group 0 to non group 0 individuals. The CABO process improves patient safety by reducing the risk of wrong blood in tube.

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AN INTEGRATED PROCESS TO PROVIDE BC EMERGENCY HEALTH SERVICES WITH GROUP 0 EMERGENCY SUPPLY RED BLOOD CELLS

***Feenstra, S.[1], Polson, K.[2], Mah, H.[1], Wand, R.[2], Roland, K.[1,4], Vu, E.[2,3,4] and Chipperfield, K.[1, 4]**

[1]Transfusion Medicine (TM), Vancouver Coastal Health (VCH); [2]British Columbia Emergency Health Services (BCEHS), BCAS Provincial AirEvac & Critical Care Operations; [3]Departments of Emergency and Critical Care Medicine; [4]Faculty of Medicine, University of British Columbia

The timely provision of red blood cells (RBC) during critical care transport can significantly impact the clinical outcomes. Early resuscitation with blood components is quickly emerging as best practice in select trauma and medical patients. However, to ensure an adequate supply of RBC's is available to clinically manage critically ill patients during transport can often be a logistical challenge; either the referring site inventory is depleted during the event or due to remoteness, the supply is non-existent. To promote best clinical practice and ensure effective RBC utilization, BCEHS approached VCH TM to develop a process for urgent access by a Critical Care Paramedic (CCP) to Group 0 Emergency Supply RBC's (ESRBC's).

The purpose of this project was to create an integrated system thal would facilitate timely access to ESRBC supply. Considerations for implementation included activation criteria, medical oversight and training, mode of transport (air, ground or water), geography and environmental conditions, validated transport containers, time constraints and location of evacuation.

Adherence to provincial and national transfusion standards was a priority. To assess the feasibility of a process that would ensure patient and product safety, a tracer audit was performed to ensure compliance with the applicable standards was achievable. In addition, a horizontal audit was performed to determine compliance with VCH transfusion administration policies and procedures.

As a result it was determined that through collaboration, an integrated process was attainable. The result is a protocol and checklist that improves patient care, supports an efficient use of provincial ESRBC resources and is compliant with the regulatory standards. As well, it provided opportunity for enhanced training in safe transfusion practice for the CCP' s. This pilot program was initiated December 18, 2013.

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INHERITED COAGULOPATHY AND HEMOGLOBINOPATHY INFORMATION PORTAL (iCHIP): IMPROVING CLINICAL OUTCOMES FOR PATIENTS WITH BLOOD DISORDERS AND OPTIMIZING FACTOR PRODUCT UTILIZATION

Jack Michaan, Sheazin Premji, Cecilia Li, Jimmy Chan, Tim Lam, Irene Pickell, *Ruby Syropiatko and Vasco Vieira-Ribeiro

BC Provincial Blood Coordinating Office

Purpose: improve clinical outcomes for patients with inherited bleeding and red cell disorders by developing an application to store health care information and provide an accurate picture of blood and blood product utilization in the hospital and the home setting.

Background: Blood and blood product utilization by British Columbians with genetic blood disorders is over $30 million (2012/13) with much of the product transfused at home. Using non-integrated data systems in the clinical setting may not optimally support patient care, analysis of utilization trends nor allow patients to provide real-time data on health issues and home transfusions. To address this, the BC Inherited Bleeding and Red Cell Disorders Services, under the BC Provincial Blood Coordinating Office (PBCO) developed the Inherited Coagulopathy and Hemoglobinopathy Information Portal (iCHIP) Patient Home Module (PHM) & Clinic Module (CM).

Method: iCHIP development required reviewing the data systems currently used and requirements gathering from clinical & patient stakeholders. The goal was to create an application to improve access to longitudinal health records, provide patients with secure access to their health information, merge records of bleeding episodes to product use (home and hospital setting), and capture transfusion data in real-time. A prototype system was used to validate the application throughout the development process.

Results: iCHIP CM was released for use by the Pediatric and Adult Inherited Bleeding and Red Cell Disorders Programs in August 2013 at BC Children's Hospital and St Paul's Hospital, respectively. To date, approximately 500 patients have their health information inputted into the Clinic Module. iCHIP PHM was released for use by patients with inherited coagulopathies on home factor product therapy in December 2013. The PBCO works with the provincial program sites to continually make improvements on iCHIP. Ultimately, this will help improve patient care delivery, care outcomes & provide information on home factor utilization.

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AVAILABILITY OF KELL NEGATIVE UNITS FOR TRANSFUSION TO FEMALES UNDER AGE 45

***Goldman, M.[1], Lane, D.[2], Gill, B.[3], Fransishyn, K.[2] and Fallis, B.[2]**

Canadian Blood Services, [1]Ottawa, [2]Winnipeg, [3]Calgary

Background: Kell alloimmunization may result in HDFN, and many European countries routinely use K- or K matched RBCs for females < 45.

Methods: We evaluated the number of CBS active donors (donated in last 2 years) phenotyped once (unconfirmed) or twice (confirmed, printed on label) for K using data warehouse, and the % of RBCs transfused to females < 45 in Manitoba in 2011 and 2012, using Traceline.

Results: Nationally, as of 11/2013, 23% of 606,086 active donors were known to be K - (14% unconfirmed, 9% confirmed): 42% in 0-, 35% in A-, 34% in B- and 19% in AB- donors; and 22% in O+, 19% in A+, 14% in B+, and 16% in AB+ donors. In Manitoba, females < 45 accounted for 8.3% of patients receiving 7.1% of RBCs in 2011; and 8.8% of patients receiving 8.0% of RBCs in 2012.

Conclusions: Due to mass phenotyping, 23% of active donors are known to be K-, well distributed across ABO and RhD groups. Approximately 8% of RBC units are transfused to females < 45. As more K- donors have a second typing done, and K-phenotyping appears on the RBC label, it should be possible to provide K- units to younger women for routine, nonurgent transfusions.

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CROSSMACH TO TRANSFUSION RATIO AND ITS ROLE AS AN INDICATOR FOR GOOD TRANSFUSION PRACTICE

***Hessah S. Alsulami, Essam Alghamdi and Ahmad Alharbi**

Alymamah Hospital, Riyadh, Saudi Arabia

Introduction: Crossmatch to transfusion ratio (C:T ratio) is the number of crossmatch tests divided by the number of transfused units. It is often used as a quality indicator of blood utilization. A high values (≥2) indicate over ordering by physicians.

Methods: This retrospective study was carried out in Alymamah hospital, one of the biggest maternity hospital in Riyadh. Our data included monthly number of crossmatched tests, number of transfused units with the initial hemoglobin levels, as well as, the diagnosis for the transfused patients which was retrieved from the blood bank records from 2012 to 2013. According to the monthly C:T ratio value, two groups were generated; accepted C:T ratio and unaccepted C:T ratio. Then, again the data from the accepted C:T ratio group was further analyzed for initial hemoglobin level, number of transfused units, and reasons for transfusion.

Results: During the period from November 2012 to October 2013, a total of 3262 crossmatching tests were done, and 1363 PRBC units were transfused with an over-all C:T ratio of 2.39, which is kind of acceptable. We also looked at the monthly C:T ratio and found that, 9 out of 12 months had C:T ratio within accepted limit (≤2). However, when the data from those months was analyzed, we found that 501 patients were transfused. Fifty eight percent (58.6%) of them had a hemoglobin level ≥ 10g/dl, only 2% of them had cardiac or pulmonary diseases but for the rest, the only documented reason for transfusion was anemia and active labor. Eighty percent (80%) of those with hemoglobin level ≥ 10g/dl were transfused with 2 units of PRBCs, 15% with 1 unit, 3% with 3units, and 2% with 4-5 units respectively.

Conclusion: Acceptable value for C:T ratio is ≤2, which means that the physicians transfused the amount of blood they ordered to their patients. On other hand, it is a quantity blood utilization indicator and does not provide any idea whether these physicians are following the transfusion guidelines or not.

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AUDIT OF TRAUMA EXSANGUINATION PROTOCOL FOR MANAGEMENT OF TRAUMA PATIENTS WITH MASSIVE BLEEDING

***Khan, E., Chipperfield, K. and Roland, K.**

Vancouver General Hospital and University of British Columbia, Vancouver, BC

Purpose: Retrospective studies have demonstrated improved survival in trauma patients who receive early Frozen Plasma (FP) in 1:1 ratio to Red Blood Cells (RBC). Based on this literature a Trauma Exsanguination protocol (TEP) was implemented at our hospital in August 2011 in which 1:1 ratio of FP:RBC is issued by the Transfusion Medicine Service (TMS) in validated transport coolers. This audit aims to review two years of experience with the TEP.

Methods: This was a retrospective review of TEP forms, patient charts and the hospital laboratory information system for all TEP activations at Vancouver General Hospital between August 2011 and Augut 2013.

Results: There were 41 TEP activations during the study period. The most common type of injury requiring TEP activation was motor vehicle accident (28%). Predetermined criteria for activating the TEP was met in 58% of cases. Patients were transfused on average 15 units of RBC and 6 units of FP. The average ratio of FP:RBC transfused in 24 hours was 1:3. The average INR upon arrival was 1:4; the average INR at 24 hours was 1:2. The average turn around time from TEP activation 10 the release of the first cooler of blood was 15 minutes. The average unused products were 8 RBC and 5 FP per patient; most of these units could be returned into TMS inventory.

Conclusion: Issuing blood in 1:1 ratio does not necessarily ensure that patients are transfused in 1:1 ratio; contributing factors will need to be explored. The predetermined criteria for activating the TEP at our hospital are often not met and therefore should be reviewed, and re-education offered. Although a significant number of RBC and FP units issued are ultimately not transfused, wastage rates can remain low if attention is made to proper storage and transport practices.

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UTILISATION DES PRODUITS SANGUINS LABILES EN CHIRURGIE CARDIAQUE

***Belleau, A., Ouellet, B. et Ouellet, P.**

CHU de Québec

Le but de l'étude était de déterminer l'utilisation versus la préparation des produits sanguins labiles lors des chirurgies cardiaques pédiatriques. La clientèle incluait les enfants de 0 à 17 ans ayant subi une chirurgie cardiaque au CHUL en 2011. Trace Line nous a permis d'extraire les volumes préparés (VP) en ml versus les volumes transfusés (VT). Notre protocole est scindé en 3 groupes d'âge : < 4 mois (G1-), 4 mois à 5 ans (G2-) et 6 à 17 ans (G3-), subdivisé en intervention avec circulation extra corporelle(CEC) et sans CEC. Les volumes à préparer sont prévus pour la période opératoire (BOP) et la période post opératoire (pour 24 h à l'unité des soins intensifs pédiatrique (USIP)). 40 interventions sans CEC pour 35 patients : pour tous ces groupes, 2 patients ont reçu une transfusion (tx) de culots au BOP, 1 patient a reçu une tx à l'USIP et 1 à l'UNN.

Le tableau suivant résume les données reliées aux interventions avec CEC.

Avec CEC	Nb de patients/ Nb de chirurgies	VP/VT (rapport) (BOP)	VP/VT (rapport) (USIP)
< 4 mois	21/21	19175/8755 (2.2)	8450/1285 (6.5)
4 mois à 5 ans	32/34	27575/14866 (1.85)	11650/1365 (8.5)
6 à 17 ans	16/16	11050/975 (11.3)	5850/975 (6)

Pour le G1- : 8 patients ont reçu du sang au BOP, 5 à l'USIP en plus du volume d'amorce. Pour le G2- : le culot d'amorce ainsi que 7 patients tx au BOP et 6 à l'USIP. De ceux-ci, 5 patients subissaient une procédure de Glenn. Pour le G3-: 3 patients ont reçu un culot au BOP, 3 à USIP. Pour cette clientèle, sauf exception, l'amorçage de la pompe s'effectue avec de l'albumine. Il a été difficile de trouver des statistiques comparatives (Schmotzer et al.Transfusion 2010 : 50 :861-67 : Meyer et al.Transfusion vol 53 Suppl S42-020D : 33A). Nous avons opté pour suivre les mêmes paramètres.

Conclusion : La mise en place d'un meilleur système des besoins des chirurgies nous permettra d'appliquer le système typage-dépistage, de permettre le transfert des unités non utilisées vers l'USIP, d'établir un nouveau rapport VP/VT qui pourra servir de mode de surveillance.

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MASSIVE TRANSFUSION IN PATIENTS WITH ACUTE UPPER GI BLEEDING: A RETROSPECTIVE CHART REVIEW

Jiheon Song[1,2], Samir C. Grever[3], Michelle Sholzberg[3], Errol Colak[4] and *Katerina Pavenski[2]

[1]School of Medicine University College Cork; [2]Department of Laboratory Medicine, St Michael's Hospital; [3]Department of Medicine, St Michael's Hospital; [4]Department of Medical Imaging, St. Michael's Hospital

Acute upper gastrointestinal bleeding (AUGIB) often necessitates transfusion. During massive hemorrhage, blood products are usually delivered at a pre-defined ratio, on the basis of protocols designed for trauma. This is antithetical to literature that has shown that liberal transfusion in AUGIB may be associated with worse outcomes. A single-centre retrospective study was conducted of all patients with AUGIB and who had massive transfusion protocol (MTP) activation. Data on hemostatic derangements, blood product requirements and clinicat outcomes were recorded.

Results: AUGIB accounted for 8% of all MTP activations. Causes of AUGIB were ulcer (65%), varices (59%) and others (12%). Coagulopathy was observed in 47% of the patients. Survival rate to discharge was 47%.

Conclusions: Individuals with AUGIB where MTP is invoked have a high mortality rate. A significant comorbidity, cirrhosis, was present in 53% of AUGIB patients with MTP activation. Also, those patients with cirrhosis showed more coagulopathy (50%) than those without cirrhosis (11%), as indicated by INR on admission.

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USE OF PROTHROMBIN COMPLEX CONCENTRATES IN THE CHAUDIERE-APPALACHES REGION 12

Chloë Bogaty, Carolle Breton, Johanne Laliberté and *Danièle Marceau

Introduction: Prothrombin complex concentrates (PCCs) are used in the rapid reversal of warfarin and all other vitamin K deficiencies in patients with a major bleed or who require an urgent procedure. The Local Advisory Committee on Transfusion Medicine standardised a protocol for the use of PCCs, outlining the indications and contra-indications for their use.

Methods: The charts of all patients having received PCCs between a two year period at the 4 hospital centers of region 12 were reviewed to evaluate the respect of guidelines, adherence to the protocol, and the clinical outcome.

Results: From Nov. 2009 to Nov. 2011, 129 patients were treated with PCCs: 97 (75%) in the emergency department, 14 (11%) in the intensive care unit, 16 (12%) on medical or surgical floors, and 2 (1.5%) in the operating room. 116 prescriptions (90%) respected the guidelines; among the extra, 4 (3%) were for elective surgeries and 3 (2.3%) for an elevated INR without evidence of bleeding. One patient did not have an INR prior to administration, 10 (7%) did not have a repeat INR 30-minutes post-administration, and Vitamin K was omitted in 10 cases (7%). The prescription is based on weight: 120 (93%) received the correct dose, 3 (2.3%) received too high a dose, and 5 (3.8%) received too low a dose. The clinical outcome was favourable in 94 cases (73%), with rapid reversal of the INR and eventual discharge from hospital. 22 (17%) died during hospitalization and 10 (8%) were lost to follow-up due to transfer to a neurosurgical facility. The INR was less than 1.4 in 86% of cases 30-minutes post-administration. 6 (4.6%) thromboembolic complications occurred: two pulmonary embolisms, two deep vein thrombosis, one stroke, one disseminated intravascular coagulation. Five had resumed warfarin, but none had a therapeutic INR.

Conclusion: Our review demonstrates that the majority of physicians correctly apply the recommended practice guidelines for the use of PCCs, that these are effective at rapidly reversing the INR and are associated with favourable clinical outcomes in the majority of cases. However, thromboembolic events are frequent, emphasizing that anticoagulation therapy should be resumed quickly once clinically appropriate.

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IMPACT OF UPDATED LETTER FOR DONORS WITH LOW HEMOGLOBIN

Uzicanin, S., *O'Brien, S. and Goldman, M.

Background: In July 2013, Canadian Blood Services began distributing a revised letter to all donors deferred for low hemoglobin (Hb) (<125g/L) (previously, females with Hb 110-I24g/L did not receive letter). Pre- and post-implementation electronic surveys assessed donor understanding and reaction to the letter, and impact of notification on actions taken.

Methods: 5,5I6 female donors randomly selected pre and post-implementation of the letter were invited by e-mail to participate in an electronic survey (response rate 39.7%).

Results:

Female Donors	Pre-imp. (N=1024)	Post-imp. (N=1165)	p-value
Received revised letter	20.1%	41.7%	<0.0001
Visited a doctor as a result of low Hb	20.5%	23.6%	0.0499
Action taken* :			
Started multivitamin + iron	10.9%	11.5%	0.6220
Started iron pills	23.7%	23.3%	0.8192
Increased iron in diet	37.6%	35.5%	0.2895
Important to tell donors if they have insufficient iron to donate	97.1%	97.2%	0.8834

*Donors who received letter took action: 76.6% pre-imp., 68.7% post-imp.

Conclusions: Although expanding distribution of the letter increased the percentage who remembered receiving it, it was still less than half, suggesting lack of impact and/or incomplete distribution. More than one method is needed to motivate donors to take action for low Hb.

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ADMINISTRATION OF INTRAVENOUS IRON IN PREOPERATIVE PATIENTS WITH ANEMIA

***Madiha Naseem[1,2], Katerina Pavenski[1,2], Katherine Luke[1], John Freedman[1,2], ONTraC Program**

[1]Department of Laboratory Medicine, St. Michael's Hospital; [2]University of Toronto

Background: Patients undergoing surgery often present with anemia. Numerous studies have shown that perioperative anemia and RBC transfusions are associated with significant morbidity and mortality. Rapid improvement in hemoglobin levels can be achieved with intravenous (IV) iron and has been shown to reduce perioperative RBC transfusions. Use of pre-operative IV iron is however inconsistent. Anecdotally, main barriers to IV iron are obtaining drug coverage and securing an infusion appointment prior to surgery.

Purpose: The aims of this study were to 1) compare use of pre-operative IV iron across ONTraC hospitals in Ontario and 2) examine the barriers to preoperative IV iron administration.

Methods: At SMH, we conducted a retrospective chart review of preoperative patients who had iron deficiency anemia and were referred for IV iron. Data were also obtained from the ONTraC records from January 1st, 2011 till December 31st, 2012.

Results: Table 1 outlines the percentage of patients who received IV iron at our and other sites. Data collection on barriers at our hospital is ongoing.

Table 1: Comparing percent IV Iron usage in pre-operative patients between St. Michael's Hospital (SMH) and all other sites in Ontario

	2011		2012	
Operation	All sites	SMH	All sites	SMH
1 knee	2.9	0	2.7	1.9
1 hip	2.7	0	3.1	0
CABG	0.8	0	1.0	0
Prostate	0.5	0	0.3	0

Conclusions: Only small percentage of patients received IV iron, despite its known efficacy. Barriers to perioperative IV iron infusion should be studied and addressed.

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SURGICAL PREOPERATIVE LEAD TIME AND UNDIAGNOSED ANEMIA

Veronika Pulley

Windsor Regional Hospital (WRH)/ ONTraC

Purpose: Research indicates that undiagnosed anemia is widespread among the general population & negatively impacts patient outcomes following surgical procedures. Timely intervention leads to the optimization of hemoglobin (Hb) levels & decreases the need for post-operative blood transfusion & associated risks. Positive results can occur if there is sufficient preoperative lead-time to allow for strategies that improve Hb. Research shows that patients with Hb levels between 100 -130 g/L pre-operatively often receive a blood transfusion postoperatively. The Ontario Nurse Transfusion Coordinators (ONTraC) developed Patient Slood Management (PSM) programs in 25 Ontario hospitals. The goal was to reduce allogeneic red cell use & improve patient access to transfusion alternatives. Initially, the program focused on defined targeted procedures, including knee replacement surgery with hip replacement surgery added in 2007.

Methos: In summer 2012, the WRH ONTraC Coordinator initiated a Blood Clinic. This provided greater lead-time & allowed patients to access strategies before preadmission visits in order to improve Hb pre-operatively. A consult request with laboratory results 15 received as early as 2 - 3 months prior to surgery. Patients are scheduled for an initial visit with the Coordlnator to: (1) review Hb & ferritin results, (2) receive an information package & (3) education on PSM strategy(s) to improve Hb. The patient & healthcare team determine which strategy(s) to implement. Preadmission visits are scheduled 3 weeks prior to surgery where Hb levels are reassessed. If Hb is not above 130 g/L, Erythropoietin can still be prescribed.

Results: Approximately 95% of patients seen had improved Hb by preadmission. Early intervention(s) include diet counseling, adding iron rich foods, & using oral iron supplements. The WRH ON TraC data showed over a 6-month period (Feb 1- July 31, 2013) a decrease in transfusion rates for hip replacement surgery from 38% (2012) to 16% (2013).

Conclusion: Early intervention through the PBM program & Blood Clinic has proven promising in improving patient outcomes. Administration 15 reviewing the feasibility of continuing & extending these strategies beyond joint replacement surgery.

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ANTI-A, ANTI-B TITER IN PLATELETPHERESIS –IMPLEMENTATION OF A SIMPLE TEST TO MEET THE NEEDS OF OUR CLIENTS

***France Bernier, Sylvie Thibault and Martin Chicoine**

Héma-Québec, Montréal, QC

Anti-A and Anti-B (anti-A,B) are found in plateletpheresis (PLTPH) at a different titer in group O donors. If transfused to Group A patients, they may induce adverse reaction.

Purpose: Implement a method to determine the titer of anti-A,B in samples from Group O PLTPH donors (GrO PLTPH Do) and label the products as « High Titer Not Detected » (HTND).

Methods: A validation study was conducted on GrO PLTPH Do to identify the titer of our donor population. Using 1 ml of plasma, a serial dilution was prepared in saline (1/1, 1/2, 1/4, … 1/512). A pool of A1 and B red cells, prepared at 2% (#cat 17318, Diagast, Loos France) mixed at a ratio of 1:1, was used. Two drops of the diluted plasma and 2 drops of the red cell pool were mixed together, incubated at room temperature and then centrifuged. The titer was determined as the highest dilution showing a 1+ positive result. Following the validation, the cut-off titer was determined and the test was implemented using a direct dilution.

Results: The antibody titer was determined on 115 GrO PLTPH Do. The titer distribution analysis showed that most of our donors were ranging between 1/16 and 1/128 (14% at 1/16, 26% at 1/32, 34% at 1/64, and 19% at 1/128). Héma-Québec, with the approval of the Provincial Committee of Blood Bank Directors, proceeded with the labeling of group O PLTPH as HTND, on donations with negative results when tested at a dilution of 1/128. The test was introduced in the daily routine mid-May 2013. From May 2013 to January 2014, 6,500 donations were tested and 4,519 (70%) were labeled as HTND. Prior to the implementation of this test, 54.6% of all PLTPH distributed to clients were group A and 35.8% were group O. After the introduction of the test, the rate decreased to 53.4% for group A and increased to 38.8% for group O. To date, 21% of the demand for Group O PLTPH is with the term HTND.

Conclusion: A simple method was developed and implemented in May 2013 to identify low titer anti-A,B Group O PTLPH. Since then, we have seen an increased demand for Group O PLTPH and a decreased demand for Group A PLTPH. This indicates that hospitals are now transfusing Group O HTND non-isogroup.

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PLATELET INVENTORY OPTIMIZATION

***Joanna McCarthy, William Martin* and Dr. Shabani-Rad**

Purpose: To develop a Platelet Inventory Management Optimization plan at CLS. In 2012 CLS had an average platelet dose discard rate of 22%.

Methods: The daily usage of the platelets at each CLS site (5) was determined. Transport of the products within the city was determined to be an issue. Ordering and recycling triggers were modified to improve distribution of product. A delivery schedule and collection day specific standing orders were created with CBS. The new deliveries used existing courier runs. These strategies eliminated 2-way transport, and allowed for earlier recycling of the doses to the central TM service.

Results: In 2013 the discard rate for platelets dropped to an average of 9%. In 2012 CLS expired 302 apheresis and 1294 pools, in 2013 we expired 227 apheresis and 464 pools; when the cost of these components is considered 619$ for apheresis and 286$ for pools (Blood Easy 2011), this decrease represents a savings of approx. 284,000$. A cost savings was also realized at CBS in the average monthly pool production, which was reduced by approx. 240 pools per month at a potential cost savings of 828,000$ per year. Overall the potential cost savings would be equivalent to 1,112,000 $ per year.

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ONE TOO MANY PLATELET TRANSFUSIONS: ARE WE MISSING PLATELET REFRACTORINESS?

Ian Marie Lano[1], Sheila Anthony[2] and *Karen L. Dallas[1,2]

[1]University of Saskachewan; [2]Saskatoon Health Region

Purpose: The goal of this retrospective, observational quality improvement study is to identify patients who may be alloimmunized to platelets and thus not receiving proper clinical benefit from unit after unit of un-matched platelets. Continued transfusion of these patients (daily or multiple times par day in some cases!) does not create any meaningful increase in their platelet counts, puts them at increased risk for transfusion reactions, and placed a significant additional cost on our already over-burdened healthcare systems. If we could identify a simple and effective strategy to alert both the laboratory and the clinical teams to possibly refractory patients earlier in their course of treatment, management could be markedly improved.

Melhods: Retrospective, Observational Chart review gathering one years' worth of transfused patients in Saskatoon. (2013) We will look at patients who've received more then one unit of platelets only. Of those patients, we will note the number and frequency of transfused units and well as the pre- and post-transfusion platelet counts. Patients who were investigated for refractorieness will be analyzed only for the time period prior to identification of refractoriness. If any transfusion reaction data is available in our system, those will be noted as well.

Conclusions: While the results are still being analyzed fully right now, the important conclusions thus far are several:

1. The majority of patients who are transfused platelets in Saskatoon do not have post transfusion CBC's performed.

2. Many patients received platelets on a daily basis with no investigation for refractoriness despite variable (even negative) increments.

Future directions: Once data is fully analyzed (will be complete within 1-2 more weaks), We will create a laboratory algorithm and meet with clinicians to discuss a clinical algoritm as well.

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PERFORMING A RETROSPECTIVE PLASMA UTILIZATION REVIEW

Mueller, D., Vandevoorde, J. and Fraser, M. D.

Health Authority

Purpose: The Fraser Health Transfusion Medicine Laboratory (TML) recognized the need for an audit of plasma transfusion practices; however, we had relatively little experience in conducting an audit of multiple hospitals within a single health authority. Our goal was to determine if the plasma transfused was clinically indicated, dosed appropriately and if an alternative therapy, such as prothrombin complex concentrate, may have been warranted. Fraser Health TML pro-actively screens plasma requests by reviewing the patient's coagulation results and the clinical indication provided in the order entry system; however, the latter is not always accurate or sufficiently detailed and we wanted to validate the assumption that prescreening prevented unnecessary plasma transfusion.

Methods: Plasma use over a 6-month period was reviewed at the four largest facilities in Fraser Health. The data elements required to complete this review were identified and included items su ch as comparison of the clinical situation to the existing TML screening criteria, patient weight, and plasma dose. This information was obtained from a review of the laboratory records, electronic patient charts and paper charts where required.

Results: The 6-month study period included review of 526 patient charts relating to transfusion of 2547 plasma units. During the course of this review, the data elements captured increased from seven to over twenty-five, significantly complicating our ability to collect and review the data and complete this review in a timely manner.

Discussion: Our goal was to determine if plasma was clinically indicated, dosed appropriately and if an alternative therapy may have been warranted; however, we also learned how to approach future utilization reviews using tools within our laboratory information system, our research department and our electronic medical records. The template modified as a result of our experience will serve as our template for future utilization reviews.

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PROJET D'AUTOSUFFISANCE EN IMMUNOGLOBULINE

***Daniel Vinet**

Héma-Québec, Montréal, QC

Dans le cadre de son plan stratégique 2012-2015, Héma-Québec s'est donnée comme mission d'accroître son taux d'autosuffisance en immunoglobuline (Iglv) issue de plasma humain. Actuellement, seulement 13 % des Iglv utilisés pour soigner les malades du Québec proviennent du plasma prélevé par Héma-Québec. Le reste, soit 87 % du plasma requis pour nos besoins est acheté sur le marché américain.

Méthodologie : La première étape du plan d'autosuffisance prévoyait la mise en place d'un centre de donneurs de plasma non rémunérés à Trois-Rivières. Ce projet pilote devait nous permettre de vérifier deux hypothèses :

1. Que la population serait prête à s'engager dans un programme de don de plasma non rémunéré.

2. Que nous pouvions prélever le plasma pour fractionnement à un coût compétitif par rapport au plasma acheté sur le marché américain.

Par ailleurs, la baisse marquée de la demande de culots globulaires et de plaquettes des centres hospitaliers, 5,2 % et 3,2 % respectivement pour les 11 premiers mois de l'exercice financier, nous amène à développer de nouvelles lignes de produits pour contrer cette baisse d'activités dans nos lignes de produits traditionnelles.

Objectifs du projet pilote :

1. Recruter 1 500 donneurs de plasma dans la région de Trois-Rivières.

2. Atteindre une fréquence de dons annuelle de 8 et un don moyen de 750 ml par don.

3. Produire le plasma de fractionnement à un coût compétitif.

Résultats : Après trois mois complets depuis la mise en service du Salon des donneurs de plasma PLASMAVIE de Trois-Rivières, nous avons accueilli 542 donneurs qui ont fait en moyenne 2,26 dons. Le volume moyen prélevé par don est de près de 700 litres et nos coûts de prélèvements s'approchent de notre objectif de départ qui était de produire à coûts compétitifs. Enfin, l'accueil de la population de Trois-Rivières a été au-delà de nos attentes.

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INTRAVENOUS IMMUNE GLOBULIN (IVIG) UTILIZATION AUDIT

***Young, L.[1], Traore, A. N.[2] and Gagliardi, K.[1]**

[1]Ontario Regional Blood Coordinating Network; [2]McMaster Transfusion Research Program

Purpose: The objectives of the 2012 IVIG audit and practice survey were: to determine the clinical indications for which IVIG is being used compared to the 2007 audit; and to ascertain compliance with 2012 Ontario IVIG Utilization Management strategy introduced in April 2012, that included implementation of a standard request form.

Methods: An audit tool designed to collect IVIG data, accessible through the Transfusion Ontario website, was provided to hospital staff. Training webinars were conducted prior to the data collection. During a three month period, from September 5 2012 to November 30 2012, non-nominal patient data, grams of IVIG used, ordering physician specialty, primary diagnosis and clinical indication were collected. A survey circulated to 139 hospitals that infuse IVIG was conducted subsequent to the audit to document strategy implementation.

Results: A total of 61 hospitals participated, including 39 that participated in 2007 and/or used 1% or greater of total IVIG used in 2011-12 and 22 volunteer sites. During the audit period, 2,446 patients, 6,442 infusions were recorded and 301,298.4 grams utilized. Results showed 87% of requests conformed to Ontario guidelines, and 13% were for clinical indications not on the guidelines. Shipments of IVIG to Ontario hospitals during 2012-13 were -1.4% compared to 2011-12, a decrease for the first time in 10 years. Shipments in 2013-14 increased. Response rate to the survey was 59%; 93% of respondents had adopted the standard request form.

Conclusion: Shipments of IVIG to Ontario in 2012-13 decreased. Most hospitals implemented the standard request form, part of the IVIG strategy. Recommendations based on audit results focus on compliance with guidelines and screening requests to encourage evidence based ordering practices. Although the audit was labor intensive for hospitals, it provided useful data on the impact of ORBCoN initiatives to improve IVIG utilization since 2007. IVIG Audit Utilization Laurie Young layoung@ mcmaster.

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BC INTRAVENOUS IMMUNE GLOBULIN NEUROMUSCULAR NEUROLOGY PROGRAM – ONE YEAR LATER

Gillian Gibson[1], Kristine Chapman[1], Russell Mosewich[2], Peter Boulton[3], Tanya Petraszko[4] Cecilia Li[4], Sheazin Premji[4], Susanna Darnel[4], Anita Gee[4] and Jennifer Danielson[4]

[1]Vancouver Coastal Health; [2]Interior Health; [3]Island Health; [4]BC Provincial Blood Coordinating Office

Purpose: To achieve optimal use of intravenous immune globulin (IVIg) for patients with neuromuscular (NM) conditions by engaging provincial leaders & stakeholders through a quality improvement approach.

Background: IVIg is an expensive possibly life-saving blood product, costing British Columbia ~$34M in 2012/13. Neurology conditions account for ~36% of BC IVIg, of which ~95% is for NM. Utilization data showed neurology as the second-fastest growing specialty user with the highest mean IVIg dosage/patient/year. To address increases & support optimal care, the BC Provincial Blood Coordinating Office (PBCO), under the Provincial Health Services Authority, began collaborating with a group of BC NM Neurologists in 2011. BC NM specialists recognized an opportunity to develop a provincial program to manage patient care & improve IVIg utilization.

Method: An IVIg NM provincial program was implemented in January 2013 with the release of a toolkit & educational video for clinical & transfusion medicine areas which included a web-based IVIg dosing calculator, optimal dosing approaches, standardized diagnostic & treatment algorithms. A provincial Review Panel was established to review specific NM IVIg requests & outcomes.

Results: Preliminary data demonstrates an improvement in IVIg utilization. Before November 2011, the average NM IVIg dosage was 94.6 grams/patient/month and during planning & implementation periods, the average decreased to 88.4. The annual utilization growth decreased from 5.6% to 1.8% in 2012/13. Although not fully measurable, there has been recognition of enhancing peer-to-peer support through the Review Panel. The next phase of work focuses on strategies for chronic users.

Conclusion: The IVIg NM program follows the success of a BC IVIg Rheumatology program. NM conditions were less well known & additional education & support was required for clinical & laboratory staff. The NM Review Panel continues to identify opportunities with stakeholders through ongoing collaboration.

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ARE WE APPROPRIATELY USING INTRAVENOUS IMMUNOGLOBULIN IN THE SASKATOON HEALTH REGION?

***Manna, M., Karim, S., Brose, K. and Dallas, K.**

Department of Medicine, University of Saskatchewan, Saskatoon, SK

Intravenous Immunogloblulin (IVIG) is a plasma protein used in the treatment of various diseases. The cost of IVIG is approximately $50-$80 per gram. In 2005, Saskatchewan's usage of IVIG was above the national average. IVIG is currently licensed by Health Canada for use in six diseases, however, IVIG is often used for off-label indications. There are several published guidelines that outline recommended uses of IVIG. Despite these guidelines, IVIG is often used in multiple settings where there is no supporting evidence.

We aimed to evaluate adherence to published guidelines by quantifying IVIG usage in the Saskatoon Health Region (SHR). Following this, we aimed to evaluate the change in IVIG usage after introducing pre-printed order forms. We conducted a retrospective chart review of patients who received IVIG in 2011 and 2012, classifying the usage based on diagnosis and prescribing specialty. In January 2014, we introduced pre-printed IVIG order forms in the SHR outlining appropriate indications for IVIG as well as provided physician education sessions on appropriate usage of IVIG. After the order sets have been implemented for three months, we will re-evaluate the usage of IVIG in the SHR.

In 2011, 227 patients received IVIG totalling 50,528g and 2.78 million dollars. In 2012, 216 patients received IVIG totalling 63,155g and 3.49 million dollars. According to published guidelines, 36% of the total usage in 2011 and 21% of the total usage in 2012 was not indicated. The data following implementation of order forms is pending, however will be ready and analyzed in time for the 2014 conference.

In conclusion, our preliminary data analysis shows that IVIG usage in the SHR does not adhere well to guidelines. Physician education sessions and order forms outlining appropriate indications for IVIG have been introduced in the SHR to help improve adherence to guidelines. Our goal is to ensure there is appropriate resource allocation of IVIG and to reduce risks to the patient and financial costs.

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IVIG ASSOCIATED THROMBOSIS: IS IT SUCH A RARE EVENT? REPORT OF A PEDIATRIC CASE AND OF QUÉBEC HEMOVIGILANCE SYSTEM

***Joy Benadiba[1], Nancy Robitaille[1], Gilles Lambert[2] and Yves Pastore[1]**

[1]Sainte-Justine Hospital, Montréal, QC; [2]Québec Hemovigilance System, Montréal, QC

Intravenous immunoglobulin (IVIG) is frequently given in autoimmune disorder. It is prepared from plasma pooled from healthy donors and used since 1952. Over the last decade, IVIG use has dramatically increased across the world. IVIG is generally considered sale and secure with acceptable adverse events. However, rare serious adverse events (SAE) can occur. Following a case report of a pediatric patient with IVIG-associated thrombotic complication, we reviewed the incidence of SAE through reports of Québec Hemovigilance System (QHS). We found 8 reports of thrombotic events possibly related to IVIG, all but one having occurred in adults. The single pediatric case occurred in a 16-year-old female receiving IVIG for history of severe immune thrombocytopenia and menorrhagia. Ten days after a second IVIG course, she developed cerebral thrombosis. QHS data are unique as it is one of the rare national hemovigilance systems thal has included IVIG reports for almost a decade, and has collected information on quantity of IVIG given since 2007. Over a five-year period (2007 to 2011), there has been 6 182 382 grams of IVIG given, and 942 adverse events reported (15.21100 OOOg IVIG, CI14.3-15.2) in the province of Québec that hosts more than 7 Mio in habitants. Thrombosis alter IVIG is therefore a rare though serious adverse event occurring mostly in adults. Given thal QHS is based upon voluntary reports, and that such the relation between IVIG and thrombosis may be under-recognized, it is possible that the incidence of such adverse event is underestimated. We underline the importance of properly reporting IVIG SAE in order to improve hemovigilance system and study such rare events.

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SUCCESSFUL TREATMENT OF FETAL ANEMIA SECONDARY TO RH ALLOIMMUNIZATION WITH PLASMAPHORESIS AND IVIG.

Mila Smithies[1], *Michiel Van Den Hof[1], Chantale Pambrun[2], Stephen Couban[3] and Jillian Coolen[1]

1Department of Obstetrics and Gynaecology, Dalhousie University, Halifax; [2]Health Centre, Department of Pathology and Laboratory Medicine, Dalhousie University, Halifax; [3]Division of Hematology, Department of Medicine, Capital District Health Authority, Halifax

Hemolytic disease of the fetus/newborn (HDFN) remains a significant cause of perinatal morbidity and mortality in pregnancies affected by red cell alloimmunization. Fetal and newborn sequelae can be avoided with early diagnosis and treatment. The clinical management of severe early Rhesus alloimmunization remains unclear. Intrauterine transfusion (IUT) is the mainstay of treatment for fetal anemia, but it is technically challenging and less likely to be successful before a gestational age of 18 to 22 weeks. In order to avoid or delay the need for IUT, much attention has been paid to alternative treatment modalities for Rh(D) alloimmunized pregnancies, namely plasmapheresis and/or intravenous immunoglobulin (IVIG).

We report the successful antepartum management of a woman whose 4th pregnancy was complicated by severe Rh(D) alloimmunization using serial Doppler measurements of middle cerebral artery peak systolic velocity (MCA-PSV) to identify and monitor fetal anemia, as well as a combination of plasmapheresis and IVIG.

To our knowledge, this is the first reported case of correlation between MCA and plasmapheresis where a multidisciplinary approach to monitoring and treating severe early onset Rh (D) alloimmunization not only slowed the progression of fetal anemia to delay the first IUT until 25 weeks gestation, but also improved fetal anemia that had already developed.

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ANTENATAL GENOTYPING ON MATERNAL BLOOD SAMPLES- THE CBS EDMONTON EXPERIENCE

***Hannon, J. L., Barr, G. and Clarke, G.**

Canadian Blood Services, Edmonton

Purpose: Genotyping on circulating free fetal DNA (cffDNA) from maternal blood samples has supplanted fetal genotyping performed on amniotic fluid or chorionic villous in many countries due to decreased fetal risk. Assays offer a degree of accuracy >98.6% after 16 weeks gestation (except Kell).

Method: In March 2013, the Diagnostic Services Laboratory (DSL) Edmonton began to offer antenatal genotyping for patients with critical antibody titres for selected Rh and Kell antibodies. DSL freezes plasma within 48 hrs. and ships to the International Blood Group Reference Laboratory (IBGRL) in Bristol, UK. The IBGRL offers fetal genotyping for RhD, Rhc, RhE, RhC from 16 weeks gestation and K (Kell) from 20 weeks. Each request is screened by a CBS DSL physician.

Results: In 2013, 18 maternal samples (15 patients) were sent for genotyping. Two (2) repeat 28 week samples for K genotyping were requested due to concern as to whether sufficient fetal DNA was present on initial typing at 20 and 21 weeks gestation. Repeat c genotyping was requested on one sample due to difficulty in initial testing. Maternal antibodies included anti-D + C (2), anti-D (3), anti-c (4), anti-E (1) and anti-K (8). Predicted fetal phenotype (PFP) was determined based on genotype results. For the first anti-D + CPFP was RhD POS (1) (C not tested). In the other anti-D+C, the fetus inherited a RHD-CE-Ds gene from the father and was considered at risk for HDFN due to the anti-C. For anti-D, PFP was RhD POS (1), RhD NEG (1) and Unable to type (1). For anti-c, PFP was Rhc NEG (1), Rhc POS (2). For anti-E, PFP was RhE POS (1). For anti-K, PFP was K NEG (4), K POS (1), Inconclusive (1).

Conclusions: 1. cffDNA testing on maternal blood is becoming standard practice in many countries. 2. Sample processing can be done in a laboratory with a biological safety hood and skilled technologists. 3. cffDNA analysis is safer for the fetus. 4. Where the fetus is negative for the gene (6/15 or 40%) the costs of follow-up ultrasound are avoided. 5. Based on our experience, we will perform Kell genotyping once during a pregnancy at 28 weeks gestation.

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ARE YOU MY MOTHER? A BLOOD GROUP DISCREPANCY STUDY FROM AN OBSTETRICAL HOSPITAL

Cook, G. and Clarke, G.

Background: ABO discrepancies in transfusion medicine (TM) laboratories typically refer to discrepancies between forward and reverse grouping in the same patient sample. We report a case of ABO discrepancy related to maternal and neonatal blood groups and a strategy for evaluation.

Case Report: The TM laboratory received a cord blood specimen. It is TM policy to check maternal blood group and screen history and test cord sample from all RH negative or group 0 mothers, to establish RhiG eligibility or the possibility of ABO HDN. A maternal history check indicated the maternal blood group was 0 positive. Standard forward grouping on the cord sample indicated group AB Rh positive. As this combination is unlikely, a step wise evaluation was initiated.

Serological Investigation: A warm saline wash of cord cells to remove Wharton's jelly (a possible contributor to a false positive AB typing) was attempted and confirmed the ABO typing. Recollection of maternal and neonatal samples also provided confirmation of previous results. A monoallelic AB gene in the mother was considered, along with chismerism or ABO subtypes in the neonate. Prier to initiating studies to investigate these possibilities, additional pregnancy history was sought

Outcome and Conclusions: The pregnancy resulted from in vitro fertilization of a donor egg. Neither maternal nor paternal genetic blood groups were known or evaluable from the parents. Communication remains the cornerstone of efficiency and accuracy. Early communication of relevant details may have prevented recollection of samples from the mother and neonate as well as serological testing and a multitude of phone calls. New fertility measures may have an impact on blood group interpretation and add a new category to the causes of unexpected maternal/neonatal blood group discrepancies.

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EVALUATION OF PROGENIKA ID CORE XT ASSAY FOR RBC GENOTYPING

Cote, J., Whitteker, T., *Pigeau, S., Scalia, V. and Goldman, M.

Canadian Blood Services

Purpose: To verify the performance of the ID CoreXT assay for RBC genotyping, with particular emphasis on the detection of additional variant CE and Lutheran alleles which were not available with the earlier Progenika IDCore+ assay.

Method: ID Core XT uses Luminex xMAP technology and analysis software to detect SNPs in 10 RBC blood group systems. The evaluation was conducted using 24 known DNA samples which included CE variants V (RH10), VS (RH20), hrS (RH19), and hrB (RH31). The acceptance criteria for the evaluation was a >95% concordance for the 24 samples tested with known genotyping results.

Results: The predicted phenotype for all blood group systems was obtained for 23 of the 24 known samples, including the additional alleles:

Group	Variants and ISBT number
RhCE	V (RH10), hr^S (RH19), hr^B (RH31)
LU	Lu^a (LU:1) Lu^b (LU:2)

One (1) sample with a predicted MN phenotype resulted as a no call during the evaluation.

Conclusions: The evaluation of the IDcore XT assay consistently identified all alleles interrogated by the assay. The routine use of this assay will be particularly useful for transfusion support of patients with sickel cell anemia, who more frequently have variant CE alleles.

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A NEW RH NULL ALLELE IDENTIFIED IN QUEBEC

***Josée Lavoie[1], Carole Éthier[2], Josée Perreault[1] and Maryse St-Louis[1]**

[1]Héma-Québec, R&D, Québec, QC; [2]Héma-Québec, Immunohematology Reference Laboratory, Québec

Purpose of the investigation: Starting in the late 1960s, samples from members of an Rh_{null} family were extensively studied by the Canadian Red Cross (Ottawa and Montréal) using approved serology techniques to phenotype D, C, c, E, e, M, N, S, s, D, P1, K, k, Fy^a/Fy^b, Jk^a/Jk^b and Ortho Research (Raritan, New Jersey) who scored sorne antigen expression (D, C, c, E, e, LW, Ce). Two sisters and one brother were found Rh_{null} (D- C- c- E- e-). The sisters were later referred to Héma-Québec's Immunohematology Reference Laboratory and became rare blood donors.

Methods: To elucidate the molecular basis of the Rh_{null} phenotype, several DNA assays were performed (RHD, RHCE*C/c, RHCE*E, RHCE*e). Rh-associated protein gene RHAG exons were sequenced.

Results: DNA results indicated that both sisters should be D+ C+ c+ E- e+ (R1r). RHAG sequencing results showed a new homozygous missense polymorphism at position 1003G>A (Gly335Ser) which could prevent Rh antigens expression.

Conclusions: The Rh_{null} is of the regulator type because it is explained by the new RHAG allele identified. The family tree showed that the three Rhnull siblings' parents were first cousins. Both parents were found Rlr and must have been heterozygous for the new RHAG missense polymorphism. Interestingly, the father's brother married the mother's sister. No Rh_{null} were found in this branch of the family. Sorne might carry the new RHAG allele, however samples were no longer available for sequencing. We believe this new allele is destined to disappear with this generation.

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IDENTIFICATION OF ANTI-GE ANTIBODIES IN FOUR MOTHERS

Morden, J.[1], Cote, J.[1], *Pigeau, S.[1], Lane, D.[1], Skeate, R.[1], Lau, W.[1], Campeau-Fleury, L.[2], Lomas-Francis, C.[3] and Goldman, M.[1]

[1]Canadian Blood Services; [2]Health Sciences North; [3]New York Blood Centre

Purpose: To investigate panreactive perinatal samples and clinical implications of the identified anti-Ge (Gerbich).

Method: Serological methods including red cells treated with papain, trypsin, -chymotrypsin and 0.2M DTT were used to determine the anti-Ge specificity. Unlicensed antisera was available for Ge:2 and Ge:3 but not Ge:4 typing. Ge phenotype was supplemented by genotyping.

Results: All typed as Ge:-2,-3, sequencing identified the deletion of GE exon 3, consistent with the Ge:-2,-3,4 phenotype. Phenotyping identified 1 Ge:-2,-3 sibling. 2 mothers (both G1P1) formed anti-Ge2; infants were DAT+, 1 required phototherapy. 1 mother (G9P7) formed anti-Ge3; the infant was DAT+, with no clinical HDN. 1 mother (G3P3) formed anti-Ge3; the infant was DAT+ and had severe HDN with suppression of erythropoiesis.

Conclusions: Anti-Ge Ab are of variable clinical importance; anti-Ge3 in particular can cause severe HDN with prolonged anemia. Siblings should be phenotyped for Ge antigens, both to increase the donor pool of available units and to plan transfusion support for future pregnancies in females of childbearing age.

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CAN THE PRENATAL TITRE BE A CLUE FOR THE PRESENCE OF ANTI-G (Rh12)?

Ronny Chan, Jenette Goldstein, *Matilda Cheung, Yulia Lin and Lisa Merkley

Sunnybrook Health Science Centre

Purpose: The perinatal policy at Sunnybrook Health Sciences Centre recommends that a Group and Screen be tested at 12 weeks gestational age. Positive antibody screens undergo further identification and when clinically significant, antibody titration. We describe a prenatal case that was referred as a previously reported anti-D and anti-C where the prenatal antibody titre results led to the identification of an anti-G and anti-C and not anti-D and anti -C.

Methods: The Antibody Screen was tested using MTS Gel Technology. The investigation was continued using panel cells in MTS and saline tube method. Ethnic backgrounds were requested on patient and current partner. Antigen typings were performed on the patient and partner using antisera.

Results: This was a 34 year old female Gravida 6, Para 2. The patient typed as 0 Rh Negative (rr), antibody screen positive. The current partner tested 0 Rh Positive (R1Ro). Initially anti-D and anti-C were identified as previously reported. Titration studies were performed using R1R1, R2R2 and r1r cells. Titres with these 3 cells were all the same at 1:512. A higher titre would have been expected with an anti-D and anti-C on R1R1 cells due to the double dose expression of both C and D antigens. A lower titre due to anti-C would have been expected with the r1r cell. As a result, anti-G was suspected. The specimen was sent to the Canadian Blood Services (CBS) reference laboratory for confirmation. CBS Reference Laboratory confirmed that they had previously identified anti-G and anti-C in 2011. The anti-G and anti-C were confirmed in the current sample.

Conclusion: This investigation demonstrates the importance of understanding the expected reactions of an anti-D +anti-C compared to anti-G. As a result of recognizing the presence of anti -G (rather than anti-D), the patient was still a candidate for Rhlg. This case also demonstrates the need for a data base of patients with clinically significant antibodies and the value of ensuring the patient is notified of special transfusion requirements.

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ANTIBODY IDENTIFICATION BY AUTOMATED SOLID PHASE PANELS: A USEFUL ADJUNCT OR EFFECTIVE, STAND-ALONE STRATEGY?

***Rizzetto, L., Chan, K., Chipperfield, K., Danesin, P., Mah, H., Sham, L., Tapp, S., White, T. and Roland , K.**

Transfusion Medicine Service, Vancouver Coastal Health, Vancouver, BC

Purpose: Due to the labor intensive nature of antibody investigations, facilities with high volume and/or marginal technical support can benefit from automation. Solid phase antibody panels provide both high sensitivity and the ability to be used with automation. The purpose of this study was to assess whether this platform can be used alone to complete the majority of antibody investigations in a high volume facility.

Methods: Immucor manufactures three different panels: Capture-R®Ready-ID, Capture-R®Ready-ID Extend 1, and Capture-R®Ready-ID Extend II, which use the principle of solid-phase red cell adherence assay (SRCA). These three panels contain donor cells selected with varying criteria to fulfill different antibody exclusion needs. Antibody investigations were performed in a high volume facility using an automated platform (NEO and ECHO analyzers, Immucor) and a combination of the three automated panels.

Results: A total of 318 antibody investigations were performed during the study period. Results were as follows: alloantibody(ies) identified (n=226), pan-reactivity (n=57), and negative results (n=35). Of the samples in which alloantibodies were identified, 49/283 (17%) were identified using only one panel, 127/283 (45%) were identified using two panels, and 12/283 (4%) were identified using all three available panels. Samples requiring further exclusions beyond the three available panels accounted for 38/283 (13%) of the antibody investigations. Pan-reactivity in 57/283 (20%) of the antibody investigations necessitated testing by other identification methods. Overall, 188/283 (66%) of antibody investigations could be completed using only automated panels.

Conclusion: Automated panels provide an effective, stand-alone strategy to complete the majority of antibody investigations in a high volume facility. This could have implications for savings in terms of cost and labor.

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A TALE OF TWO CITIES (AND TWO PATIENTS)

Berry, B.[1], *Clarke, G.[2] and Nahirniak, S.[2]

[1]Royal Jubilee Hospital; [2]University of Alberta

Background: Several ethical, technical and logistical issues are related to finding rare donor units for patients in need. Case report: An 80 year old female with unstable angina and moderate anemia required urgent cardiac surgery. Pre-surgical serologic testing revealed an antibody to a high incidence antigen. An historical record check indicated an anti Jra had been previously identified. Urgent blood requirement led to un-orthodox identification and a hospital to hospital transfer of Jra negative red cell units collected for autologous and directed use (for infant) from a pregnant patient in another province who also had anti Jra antibodies. In the quest to find suitable units the issues and barriers included: the ethics / logistics of public appeals for rare units; the ethics / logistics of requesting blood from family members; the availability, reliability and interpretation of the monocyte monolayer assay for assessment of the antibody's clinical significance; the availability and clinical utility of stealth cells; the strategy for inventory management of the national rare donor registry and blood bank; the technical difficulties of reliably typing rare donor cells and maintaining and tracking rare typing sera and cells; the links and communication between international blood collection agencies and the means, ethics and logistics of contacting international agencies as well as transporting and accepting blood components from across our borders. Our patients were successfully treated - one required no blood, the other received 3 units originally collected as autologous units for the other, one allogeneic unit from a known Jra negative volunteer donor and one allogeneic unit collected from a family member of another patient with the same antibody. Conclusion: The process of identifying and tracking appropriate donor units for use in a setting of urgent transfusion need requires ongoing and effective communication between blood suppliers and hospital transfusion services.

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MNS TYPING PROBLEMS

***Josée Perreault[1], Carole Éthier[2], Josée Lavoie[1] and Maryse St-Louis[1]**

[1]Héma-Québec, R&D, Québec, QC; [2]Héma-Québec, Immunohematology Reference Laboratory, Québec, QC

Purpose of the investigation: Two cases were referred to Héma-Québec's Immunohematology Reference Laboratory for MNS discrepancies: Case 1 involved the S antigen and Case 2, the s antigen. Case 1: a 72 year old Caucasian male treated in oncology, Case 2: a 22 year old Black woman suffering from sickle cell disease (hip replacement).

Methods: Approved serology methods were used. The S and s phenotype was done with three sources of anti-S (Ortho, DBL and Immucor) and one source of anti-s (Lome). For molecular biology, DNA as well as RNA were tested by PCR-SSP and/or sequencing.

Results:

Results are presented in the following table:

Case	Phenotype	Molecular biology	
		DNA	RNA
1	S- (hospital) S+s+ S+s+ S-s+ Note: S- with DBL only	*MNS*03/MNS*04* *MNS*03/MNS*04* *MNS*04/MNS*04 + MNS*05*	*MNS*04/MNS*04*
2	s+ (hospital) S+s+ (hypo) S+s+	*MNS*03/MNS*03* *MNS*03/MNS*04*	*MNS*03/MNS*03* *MNS*03/MNS*04*

Conclusions: Both cases showed a discrepancy in the MNS blood group system. Case 1 S phenotype was positive with sorne reagents and negative with DBL. Case 2 had a phenotype/genotype discrepancy. DNA and RNA analyses were inconclusive. More work is still needed to elucidate these two cases.

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EVIDENCE AGAINST A SIMPLE RELATIONSHIP BETWEEN THE ABILITY OF ANTI-RBC ANTIBODIES TO INDUCE ANEMIA AND THE AMELIORATION OF PASSIVELY-ACQUIRED MURINE ITP

***Chao Ching Jen[1] and Alan H. Lazarus[1,2]**

[1]Li Ka Shing Knowledge Institute of St. Michael's Hospital; [2]Canadian Blood Services; [3]Department of Laboratory Medicine, University of Toronto; Toronto, ON

Introduction: Immune thrombocytopenia (ITP) is an autoimmune disease that is characterized by platelet destruction mediated by Fc receptor-dependent phagocytosis. A first line therapy for ITP is Rh immune globulin (anti-D). It has been speculated that anti-D works by blocking the function of the mononuclear phagocytic system (MPS) through engaging Fc receptors on macrophages, thus competing out the MPS ability to clear sensitized platelets. If this theory is correct then the ability of an anti-RBC therapeutic to ameliorate ITP could be dependent on the development of anemia. To address this, we have treated ITP mice with anti-RBC antibodies (Abs) to look for a relationship between anemia and therapeutic activity.

Methods/Results: HOD mice were treated with an anti-RBC Ab specific for murine Glycophorin A (TER119) and Abs specific for different portions of a model red cell antigen, the HOD molecule. The HOD molecule is consisted of extracellular hen-egg-lysozyme linked in tandem sequence to ovalbumin then to transmembrane human Duffyb protein. After 24 hours treatment, thrombocytopenia was induced by anti-platelet Ab. It was observed that two Duffy-specific Abs (Mima29 & CBC-512) were able to induce anemia but not ameliorate ITP. In contrast as previously found, TER119 was able to ameliorate ITP (P=0.0055); however, partial anemia was induced at this time point. To assess if TER119 has the ability to ameliorate ITP at the time point where maximal anemia was occurring (four days after administration), TER119 treated mice were injected with anti-platelet Ab after four days. No therapeutic response was observed under these conditions despite the presence of severe anemia.

Conclusion: These findings suggest that anemia is not a sufficient condition in the amelioration of ITP by the RBC specific Abs evaluated; nonetheless, while the degree of anemia does not correlate with therapeutic activity, RBC clearance mechanisms initiated by these antibodies may still play a role in their therapeutic mechanisms.

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IMPACT OF INTRAVENOUS IMMUNOGLOBULINS ON HIPPOCAMPAL PROTEOME IN 3XTG-AD MOUSE MODEL OF ALZHEIMER'S DISEASE

***Cassandra R. Goulet[1,2], Isabelle St-Amour[1,2], Renée Bazin[2] and Frédéric Calon[1]**

[1]Axe neurosciences, Centre de Recherche du CHU de Québec, QC; [2]Département de recherche et développement, HémaQuébec, Québec, QC

Introduction: As people are living longer than ever before, Alzheimer's disease (AD) is a growing public health concern that affects over 36 million people worldwide. Current approved treatments for AD offer short-term clinical benefits but fail to alter disease progression. More recently, intravenous immunoglobulin (IVIg) treatment has emerged as an alternative therapy. Despite negative results in most recent clinical trials, two AD-related IVIg trials are still in progress and preliminary data suggest benefit for cognition only in subgroups of patients. However, the mechanisms underlying the effect of IVIg remain unknown and preclinical studies are urgently needed.

Methods: Two-dimensional electrophoresis was performed to evaluate the effect of IVIg treatment on hippocampal proteome in the 3xTg-AD mouse model of AD. Mice received two weekly injections of IVIg (1,5 g/kg) for three months and were sacrificed at 16 months of age. Proteins were isolated from hippocampus and separated by isoelectric point in the first dimension and by mass in the second dimension. The spot detection, volume calculation and comparison analysis were performed using Progenesis SameSpots software.

Results: Comparing the density of spots between four IVIg-treated and untreated mice, we found that five protein spots were significantly increased and nine were decreased in the IVIg-treated group. Mass spectrometry protein identification is currently underway.

Conclusion: We generated one of the first snapshot of changes in brain protein expression occurring after IVIg treatment, thereby providing new insights into IVIg mechanisms in an AD model.

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THE EFFECT OF ADDITIVE SOLUTIONS ON RAT AND HUMAN RED BLOOD CELLS FOLLOWING BLOOD COMPONENT MANUFACTURING

Luciana da S. Cavalcante, Jason P. Acker and *Jelena L. Holovati

Background: The use of animal models is becoming an essential step in preclinical studies of blood transfusion products. Although there are multiple studies on the effects of blood banking practices on human red blood cells (RBCs). little is known about the effect that standard blood component processing has on quality of rat RBCs.

Objectives: The aim of the study was to compare rat and human RBC metabolic and membrane-related quality parameters when a buffy-coat component production method is applied.

Methods: Blood from Sprague-Dawley rats and human volunteers (n=6) was collected in CPD anticoagulant, resuspended in SAGM or AS3, and leukoreduced. In vitro quality was analyzed, including defonnability, aggregation, microvesiculation, phosphatidylserine expression,% hemolysis, ATP, 2,3-DPG, osmotic fragility, sodium and potassium concentrations.

Results: Rat RBCs had decreased deformability, membrane rigidity, aggregability and microvessiculation after component manufacturing process, when compared to human RBCs. Rat RBCs in SAGM showed higher hemolysis compared to human RBCs in SAGM (rat: 4.70% ± 0.83% vs. human: 0.34% ± 0.07%, p = 0.002). Rat RBCs in AS3 had greater deformability and rigidity than in SAGM. The number of MPs/IJL and the percentage of phosphatidylserine expression ware lower in AS3 rat RBCs than SAGM rat RBCs (p = 0.046, p=0.028, retrospectively). Hemolysis was also significantly lower in AS3 compared to SAGM (2.21% ± 0.68% vs. 0.87% ± 0.39%, p =0.028).

Conclusion: Rat RBCs significantly differ from human RBCs in metabolic and membrane-related aspects, which should be taken into account when performing preclinical transfusion studies using the rodent model. Additive solutions play an important role in RBC preservation; however SAGM, which is commonly used for human RBC banking, is associated with more significant membrane injury in rat RBCs.

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RETICULOCYTE HEMOGLOBIN EQUIVALENT (RET-He) IN THE MANAGEMENT OF PREOPERATIVE ANEMIA; IS IT USEFUL INFORMATION? A PILOT STUDY

***Donna Touchie, Matthew Henderson, Emily Goyette, Ruth Padmore and Antonio Giulivi**

The Ottawa Hospital

Purpose: Reticulocyte hemoglobin equivalent (RET-He) is a new CBC parameter available from modern CBC analyzers. The RET-He provides a direct estimate of the recent functional availability of iron into erythrocyte hemoglobin and has clinical utility in evaluating changes in iron status in renal dialysis patients. The clinical utility of RET-He in the diagnosis and management of preoperative (preop) iron deficiency has not been studied. The purpose of this study is to observe the relationship between the results of standard iron studies, hemoglobin (Hgb) levels and the RET-He in patients receiving preop intravenous (IV) iron therapy.

Methods: A retrospective study was conducted on fifty consecutive preop patients treated for iron deficiency. The results of Hgb, serum iron studies and RET-He were observed pre and post IV iron therapy.

Results: Inspection of the data using slope plots for Hgb and RET -He pre and post IV iron therapy was not informative due to the wide variation in timing of the pre and post blood work relative to the date of the IV iron therapy. The raw data was standardized as percent change and percent rate of change for the Hgb and RET -He after the 1 st IV iron infusion. The only time interval measured was between the 1 st IV iron infusion and the 1st post IV iron infusion blood work. The greatest percent and rate of increase in RET-He was observed in the samples obtained soonest (3 days) after IV infusion, and declined steadily thereafter, and with the most rapid increase seen with lower pre-infusion RET-He levels. The Hgb level did not show a positive percent and rate of increase until approximately 7 days after IV iron infusion. Linear regression model for rate of change of RET-He suggests that low pre-infusion concentrations of RET-He and higher pre-infusion concentrations of transferrin result in a greater rate of change in RET-He post infusion (P-values <0.001 and 0.028 respectively).

Conclusions: Pre-infusion RET-He (low levels) and pre-infusion transferrin (high levels) show greater rate of change in RET-He post infusion. These findings may aid in future study design for optimal preop iron therapy.

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ÉVALUATION DE L'UTILITÉ DU FLOSEAL® COMME AGENT HÉMOSTATIQUE TOPIQUE

***Carolle Breton, Johanne Laliberté, Danièle Marceau et les membres du comité de médecine transfusionnelle de la région 12**

Introduction : Les agents hémostatiques topiques (Floseal®, Tisseel VH S/D etc.) sont de plus en plus utilisés en chirurgie en tant qu'adjuvant à l'hémostase lorsque la maîtrise des saignements par ligature ou autre procédures conventionnelles est inefficace ou impraticable. Au Québec, ces produits figurent sur la liste des produits sanguins disponibles à Héma-Québec. Nous rapportons l'étude de cohorte prospective de l'utilisation de l'agent hémostatique Floseal®, à partir de son introduction au centre hospitalier régional, CSSS Alphonse Desjardins – Hôtel-Dieu de Lévis.

Méthodologie : Les membres du comité régional de médecine transfusionnelle ont recommandé une cueillette de données pour l'évaluation de l'utilité et de l'efficacité du produit Floseal®. Une fiche visuelle d'évaluation de l'efficacité du produit sur une échelle de 1 à 10 où 10 était complètement hémostatique et 1 totalement inefficace a été élaborée. Cette fiche était acheminée avec le produit à la salle d'opération, complétée par le chirurgien prescripteur et retournée au service de banque de sang à la fin de l'intervention chirurgicale.

Résultats : De l'introduction dans notre milieu en 2008 au début de 2013, 525 fiches ont été remplies avec une augmentation significative de l'utilisation au cours des années. La chirurgie vasculaire est la plus grande utilisatrice, l'orthopédie et l'ORL l'utilisant le moins. Plus fréquemment le produit est utilisé, plus la satisfaction est grande. La chirurgie vasculaire avec 37,1% (195) des utilisations évaluant le produit à 9.8 ± 0.1/10, alors qu'en chirurgie gynécologique avec 8,2% (43) la satisfaction n'est que de 8.1± 2,6/10. Le produit semble moins efficace dans certaines chirurgies : salpingectomies (satisfaction 2/10), conduit iléal (5/10), chirurgies de LEFORT (6/10), grossesses ectopiques (6/10) et amygdalectomies (6/10).

Conclusion : Cette collecte de données nous permet de conclure à l'efficacité générale du produit avec une grande satisfaction des médecins utilisateurs. Il semble y avoir certaines interventions chirurgicales où l'efficacité reste à être démontrée.

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CHANGING A LIFE, DROP BY DROP

Claudia Bédard*, Mélissa Girard, Jocelyne Dion, Isabelle Laliberté, Marc Germain and Louis Thibault

Héma-Québec, Québec City, QC; Centre hospitalier de l'Université Laval, Québec City, QC

Purpose: Ligneous conjunctivitis is a rare disease characterized by the formation of dense membranes on the palpebral conjunctiva. These membranes are formed by deposits of fibrin, a protein involved in blood clotting. Since the disease is known to be a consequence of an underlying type I plasminogen deficiency, it usually responds to the topical application of unprocessed human plasma. We herein describe the case of a young patient who did not respond to standard therapy and who was successfully treated with plasminogen concentrate eye drops, a medication which is not available in Canada.

Methods: Plasminogen concentrate was purified from ABO-compatible human plasma using a commercial affinity chromatography Lysine-Sepharose resin. Plasminogen was eluted with Epsilon-aminocaproic acid. Peak fractions of plasminogen were pooled, precipitated with 75% ammonium sulfate and dialyzed against saline solution. Following filtration, plasminogen concentrate was diluted in sterile saline to a concentration of approximately 1.5 mg/mL and stored at -35°C until its use as eye drops. The patient received one drop per hour, in each eye.

Results: A purity of 98% was obtained for the plasminogen concentrate. Patient condition rapidly improved within a few days of treatment. Three weeks following the beginning of concentrate plasminogen eye drops, complete resolution of membranes was noted and no relapse was observed. After four months of treatment, intervals between eye drops application were increased to two hours and all other medications were stopped.

Conclusions: This report presents another rare case of ligneous conjunctivitis that required treatment with topical plasminogen concentrate. Blindness has been avoided in this young patient and future observations will reveal if eye drops can be completely discontinued.

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COMPARISON OF IN VITRO PARAMETERS OF RED BLOOD CELLS WASHED WITH DEXTROSE SALINE OR RINGER'S ACETATE AND STORED IN AS-3

***Audrey Laforce-Lavoie, Annie Jacques, Mélissa Girard and Louis Thibault**

Héma-Québec, R&D, Québec City, QC

Purpose: Red blood cells (RBCs) are occasionally washed to remove plasma proteins and metabolites that accumulate in the unit during storage. The Haemonetics ACP 215 automated cell washers, recently put into use at Héma-Québec to process washed RBCs, use a solution of 0.2% dextrose in 0.9% saline as washing solution. This solution of dextrose-saline is a minimal medium that preserves the integrity of RBCs by providing a source of energy to produce ATP and maintain cell viability. Recently, it has been demonstrated that washing of platelets in neutral, calcium-free, Ringer's acetate allows good recovery of platelets and a better preservation of their in vitro functions. In this work, we compared in vitro parameters of RBCs washed using dextrose-saline and Ringer's acetate solutions.

Methods: Leukoreduced RBC units were prepared from 6 healthy volunteers and stored in AS-3. ABO-compatible RBC units were pooled and split in pair and stored at 1-6°C. On day 14, paired RBC units underwent a double-washed procedure with dextrose-saline or Ringer's acetate. After wash, RBCs were stored for 14 days and samples were taken on day 1, 3, 7, and 14 post-wash to perform in vitro analysis (glucose, lactate, potassium, ATP, hemolysis).

Results: ATP decreased gradually during storage in both washing conditions. RBCs washed with dextrose-saline show slightly higher glucose levels than those washed with Ringer's acetate. Glucose consumption remains low during storage with both solutions. Inversely, potassium levels are higher in the RBCs washed in Ringer's acetate. No differences are observed for lactate levels in RBCs washed either with dextrose-saline or Ringer's acetate. Hemolysis is slightly greater in RBCs washed with dextrose-saline solution after 7 day post-wash but always stays below the acceptable level of 0.8% in all samples.

Conclusions: Although Ringer's acetate solution seems more effective in reducing the RBC percentage of hemolysis post-wash, none of the two washing solutions seems to be more effective than the other to improve the quality of our washed RBC units.

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RED BLOOD CELLS WASHING WITH THE ACP 215 SYSTEM: A VALIDATION STUDY

***Audrey Laforce-Lavoie, Annie Jacques, Marie-Claire Chevrier, Mélissa Girard and Louis Thibault**

Héma-Québec, R&D, Québec City, QC; Héma-Québec, Reference and Stem Cell Laboratory, Montréal, QC

Purpose: The closed-system ACP-215 (Haemonetics) enables automated washing of red blood cells (RBCs) and extended post-wash storage to 14 days in SAGM solution. Few data are available regarding the in vitro quality of RBCs washed with this technology and stored in AS-3 solution. This report presents the data for the validation study of RBC stored in AS-3 after washing with the automated ACP-215 system.

Methods: 14-day AS-3 and SAGM RBCs were washed with the ACP-215 system and stored post-wash for 7 days in AS-3. One or two consecutive procedures with 1.84 L of saline solution were validated (n=48 per arm). Acceptability criteria (sterility, hematocrit (≤ 0.8 L/L), hemoglobin (≥ 35g/unit), hemolysis (< 0.8%), and IgA titers (<0.5 µg/mL)), were studied on day 7 post-wash.

Results: All units met acceptability criteria on day 7post-wash (Table I). One AS-3 RBC unit has an hemolysis percent of 0.8%. All units were sterile.

Table I: Post-wash quality parameters of 14-day RBCs*

	1 wash		2 washes	
	AS-3	**SAGM**	**AS-3**	**SAGM**
Hematocrit	0.52±0.04	0.51±0.04	0.49±0.06	0.47±0.07
Hemoglobin	55±5	50±5	52±6	48±4
Hemolysis	0.3±0.1	0.2±0.1	0.4±0.1	0.3±0.1
IgA titer†	N/A	N/A	0.02±0.01	0.02±0.01

*Results are expressed as mean±SD. †IgA titer was done on day 1 post-wash.

Conclusions: RBCs stored for 14 days before washing can efficiently be washed with the ACP-215 system and stored for up to 7 days post-wash when stored in AS-3. RBC washing with the ACP-215 system has been implemented on November 10th 2013 in our operations thus facilitating the use of washed RBCs by our hospital centers.

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HÉMOLYSE PAR RÉCHAUFFEMENT: COMPARAISON DES RÉCHAUFFE-LIQUIDES HL-90 ET HL-90-38

Thomas G. Poder[1,2], Denise Pruneau[3], Josée Dorval[3], *Louis Thibault[4], Daniel Sale Ménard[5], Jean-François Fisette[2], Suzanne K. Bédard[2], Martine Comtois[3], Lise L. Blais[6], Flavien N. Andrianary[5], Christian Bellemare[2] et Patrice Beauregard[7]

[1]CRC, CHUS; [2]UETMIS, CHUS; [3]Banque de sang, CHUS; [4]Héma-Québec; [5]GBM, CHUS; [6]Soins infirmiers, CHUS; [7]Service d'hémato-oncologie, CHUS

Peu d'informations sont disponibles dans les notices d'accompagnement émises par les manufacturiers de produits sanguins sur la température maximale de réchauffement et le risque associé d'hémolyse alors que les réchauffe-liquides offerts sur le marché dépassent fréquemment les 38 °C et atteignent parfois 43 °C. L'objectif de cette étude, réalisée dans le cadre d'un audit de qualité, est de vérifier si l'utilisation d'un réchauffe-liquide à 41,5 °C est aussi sécuritaire en termes de niveau d'hémolyse que l'utilisation d'un réchauffe-liquide à 37,5 °C. Les réchauffe-liquides utilisés sont ceux de la compagnie Smiths Medical (HL-90 et HL-90-38). La méthodologie utilisée est celle d'une étude pré-post du niveau d'hémolyse (apprécié par le niveau d'hémoglobine libre et le pourcentage d'hémolyse) suite au réchauffement du sang dans l'un des deux réchauffe-liquides. Le sang utilisé correspond à des culots globulaires de groupe AB positif de 42 jours et moins. Chaque culot est divisé en deux afin de procéder aux tests sur les deux réchauffe-liquides. Les tests ont été réalisés selon différents débits et techniques d'administration (pompe à perfusion, transfusion d'échange en néonatalogie, manchon compressif). Les résultats indiquent qu'un réchauffement à 41,5 °C ne produit pas significativement davantage d'hémolyse qu'un réchauffement à 37,5 °C. Le niveau d'hémolyse ne s'accroît pas de façon significative avec le réchauffement, ni avec la transfusion d'échange ni avec le manchon compressif. Par contre, l'utilisation d'une pompe à perfusion (Symbiq), en particulier avec un débit de 600 ml/h, augmente significativement le niveau d'hémolyse. En conclusion, un réchauffement à 41,5 °C ne compromet pas davantage la sécurité transfusionnelle qu'à 37,5 °C.

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EVALUATION OF THE COMPOGUARD AUTOMATED BLOOD COLLECTION MIXER

***Marie-Eve Allard, Annie Jacques, Marie-Eve Nolin, Claudia Bédard, Mélissa Girard and Louis Thibault**

Héma-Québec, R&D, Québec City, QC

Purpose: The selection of a blood mixer for blood collection must rely not only on its cost but also on an evaluation of its impacts on clinic logistics, the anticoagulant: blood mixing to prevent clot formation and on the precision of the volume. In this work, we compared the performance of the CompoGuard blood mixer (Fresenius Kabi) with our current instrument Sebra 1040 (Haemonetics).

Methods: The precision and accuracy were studied at 30, 50 and 75 mL/min using a 30% glycerol solution to simulate whole blood density and viscosity. The mixing efficacy was studied by pumping the glycerol solution in collection sets from three manufacturers in which the anticoagulant has been stained with 150 mM of toluidine blue. Bags were next suspended and fractionated into portions of 12 mL and the mixing efficacy was obtained by comparing optical densities (ODs) at 640 nm. Whole blood was collected from volunteers after informed consent and the presence of clotting was verified by filtering 50 mL on Whatman cellulose filters. Impacts of mixers on the staff workload, operational procedures and logistics were evaluated.

Results: Blood mixer precision was not affected by collection flow rate and was ≤99.5% and ≤97.5% for the CompoGuard and the Sebra 1040, respectively. The CompoGuard was more accurate than the Sebra 1040 mixer (≥99.3% compared to ≥92.7%). Experiments using the toluidine blue indicated an acceptable mixing of anticoagulant. However, the presence of small clots in 58% of bags collected with the CompoGuard comparatively to 8% with the Sebra 1040 indicates a weaker mixing capacity. The CompoGuard is easily adaptable to our different collection sets and its data logging capacity enables the traceability of collections.

Conclusions: The CompoGuard presents several operational benefits over the Sebra 1040 mixer. Despite mixing and weighting acceptable performance, manual mixing of blood bags after collection is strongly advisable to prevent clots formation with the CompoGuard.

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QUALIFICATION OF REFRIGERATORS/ FREEZERS

***Birch, P., Leung, D., Madana, X., Mallon, K., Hashimi, M., Burke, T., Milne, T. and McGuirl, M.**

Canadian Blood Services, Head Office, Validation Department

Objective: The qualification of Canadian Blood Services (CBS) refrigerators and freezers following current regulatory and industry requirements was described in this study. The Validation and Equipment Services department supports, monitors and improves the nationalized standardization process within CBS. In a manufacturing environment the equipment and systems which impact blood and blood components must be in a state of control this includes the qualification of all new equipment or equipment that has undergone major repairs.

Design/Materials and Methods: CBS has several hundred refrigerators and freezers in the field of various sizes from chest and upright to walk-in models and varying in price from 10K to over 300K. The Validation department has the task of developing and executing protocols for these to be reviewed by Quality Assurance, Internal Audits and Health Canada. Protocols include basic operations, alarms, electrical specification checking, temperature range mapping while empty and full of mock product plus performance under worst case scenario's of power outage and open doors.

Results: Presented here are the compilation of several refrigerator and freezer validations. Pictures of the chamber during temperature mapping, graphical results of the mapping and interesting problems encountered during validation are included. Common problems found at various sites include electrical setup of critical alarm and recorder systems was non CSA approved and improper labeling of breakers, compressors, alarm sensors and condensers.

Conclusions: The results obtained from our study demonstrate common issues encountered and shows that cooperation and buy in to the validation system is required between the users, facilities and quality systems to help stream-line the process.

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MONITORING RESIDUAL PLASMA VOLUME IN RED BLOOD CELL UNITS: A NEW TOOL FOR BLOOD PRODUCT QUALITY CONTROL

Louis Thibault, Jocelyne Dion, Sophie Dubuc, Annie Jacques, Pierre Robillard, Claudia Bédard, Mélissa Girard and Marie Joëlle de Grandmont

Héma-Québec, Québec City and Montréal, QC

Purpose: Currently no national or international standards address the content of residual plasma in red blood cell (RBC) units despite the fact that this residual plasma represents a potential risk for the transfused recipient and a loss of efficiency for blood product manufacturers. Several factors such as blood: anticoagulant ratio, centrifugation or apheresis apparatus settings, plasma extraction and additive solution addition can modulate the volume of plasma in RBCs making it difficult to estimate. In this work is presented the approach taken by Héma-Québec to monitor the volume of plasma in RBCs as part of our quality control (QC) program.

Methods: The amount of residual plasma in RBCs was established based on the immunoglobulin G (IgG), measured in the supernatant and in plasma of apheresis RBCs and whole blood derived AS-3 and SAGM RBCs (60 of each arm). In parallel, the plasma content of RBCs was also mathematically estimated based on unit volume and hematocrit minus the additive solution volume. A relationship between measured and estimated plasma volumes, obtained from a linear regression analysis, was used by QC to monitor products manufactured since June 2013.

Results: Using IgG ratio, volumes of plasma were 39 ± 12 mL, 23 ± 5 mL and 38 ± 9 mL in AS-3, SAGM and apheresis RBCs. The relationship between the plasma volume measured according to the IgG ratio and the estimated volume was R2 = 0.8583 and the following formula was established: [(((1-HCT) x RBC volume) - additive solution) - 7.1317] / 0.938. Of the 1,939 analyzed products, the average plasma was 28 ± 13 mL per unit and 95% had less than 50 mL of plasma. No difference was found between RBCs produced by our two production sites.

Conclusions: Calculation of the volume of plasma in RBCs can be easily integrated with all other activities of the CQ laboratory. Such measurements can be used as indicators of quality but may also be very useful for improving the performance and standardization of blood manufacturing.

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EXCURSION OF FROZEN PLASMA UNITS TO AMBIENT TEMPERATURE: HOW LONG BAGS CAN BE LEFT OUT OF CONTROLLED TEMPERATURE STORAGE?

***Nathalie Dussault, Marie Joëlle de Grandmont, Claudia Bédard, Mélissa Girard, Danny Brouard and Louis Thibault**

Héma-Québec, R&D, Québec City, QC

Purpose: Recently, an audit of the American Association of Blood Bank (AABB) has pointed out that there is no validation protocol to support the 30-minute period currently allowed in our organization for the labelling of frozen plasma (FP) units at room temperature (RT). Most regulatory agencies state that internal temperature of FP units should never be higher than -18°C. In this work, preliminary results for the monitoring of internal temperature of FP units exposed to RT for 60 minutes are presented.

Methods: Plasma was obtained from whole blood (WB) bags and expired apheresis donations. Plasma samples were pooled and split into empty bags (n=5 per arm) to mimic WB-derived plasma units of 322 mL (group A), 268 mL (group B) and 137 mL (Group C) as well as apheresis plasma of 462 mL (group D) and 137 mL (Group E). Following a minimal 16-hour freezing period at -25°C, the core temperature was monitored using a data logger and temperature probes positioned inside the bags during exposure at 20-24°C. Visual inspection was also performed every 10 minutes to observe plasma melting.

Results: Table 1 presents the average time required for FP units to reach the critical temperature of -18°C. According to these results, FP units reached the critical core temperature of -18°C in less than 12 minutes. As expected, low volume samples thawed more rapidly than large ones. After 20 minutes, the presence of thawed plasma was observed in the corners of the bags.

Table 1: Time needed for FP units to reach a core temperature of -18°C

Groups	A	B	C	D	E
Time (min)	11	9	4	9	3
Min-Max	7-14	7-12	1-5	4-13	1-5

Conclusion: This work suggests that the indiscriminate application of the "30-minute rule" may increase plasma core temperature above -18°C. A validation study should be planned in the near future to challenge this standard and define the maximum period of time FP units can be exposed to RT based on plasma protein activity.

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RAPID NUCLEATED RED BLOOD CELL ENUMERATION BY FLOW CYTOMETRY

Carl Simard, Marc Cloutier, Lise Bourgouin, Jocelyne Dion and Sonia Néron

Héma-Québec, R&D, Québec City, QC

The enumeration of nucleated red blood cells (NRBC) in cord blood units is a required criterion for QA in cord blood banks. Although specialized hematological analyzers exist for NRBC enumeration, manual count on blood film is still considered the gold standard. Because QA labs already have flow cytometers for CD34 enumeration, the same instrument could be used for NRBC enumeration. Based on the method developed by Tsuji at al.(Cytometry 37:291 1999), we have performed a study comparing NRBC enumeration from 34 cord blood samples by flow cytometry, on an Accuri C6, and blood film. Enumerations showed good Pearson's correlation ($r = 0.83$, 2-tailed $p < 0.0001$). Although we observed a slight, but clinically irrelevant, bias of 1.4% NRBC overestimation for flow cytometry, Bland-Aitman analysis confirmed a good agreement between the two methods. We conclude that NRBC enumeration by flow cytometry could efficiently replace manual count on blood film. This should provide a faster, easy to automate, procedure for the enumeration of NRBC. A validation study is now in progress to incorporate NRBC enumeration by flow cytometry as a routine test in Héma-Québec cord blood bank facility.

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EFFECT OF THE PRE-PROCESSING STORAGE TEMPERATURE ON THE POTENCY OF CORD BLOOD STEM CELLS.

***Marie-Ève Rhéaume[1,3], Diane Fournier[2], Marie-Claire Chevrier[2] and Renée Bazin[1,3]**

Héma-Québec, [1]Innovation R&D and [2]Reference and Stem Cell Laboratory, Montréal, QC; [3]Laval University, Québec, QC

Background: Umbilical cord blood (UCB) has been proven to be an important alternative source of hematopoietic stem cells (HSCs) mostly for pediatric patients suffering from hematologic disorders. Because of its many advantages over other sources of HSCs, such as ease of collection, less stringent HLA restrictions and lower risks of developing GVHD, the use of UCB has expanded in recent years and led to a growing number of public cord blood banks that operate under guidelines established by the regulatory organisms such as Netcord FACTor AABB. Despite the standardization of cord blood banking procedures, some parameters remain unregulated, such as the pre-processing storage temperature. At Héma-Québec's public cord blood bank, the units are kept at room temperature (RT) before being processed and cryopreserved within 48 hours after collection. Recently, a study using a mouse model of engraftment revealed that a pre-processing storage of 72 hours at room temperature might have deleterious effects on the HSC's hematopoietic reconstitution capacities. The aim of our study was to evaluate the impact of pre-processing storage temperature on HSCs, using the banking procedure in place in our cord blood bank.

Methods: UCB units were split into two smaller units and stored at 4 °C or RT until reaching 48 hours after collection before processing and freezing . The viability (flow cytometry), differentiation capacities (CFU) and in vivo hematopoietic reconstitution of Nod/SC ID gamma null (NSG) mice were evaluated after thawing the stored units.

Results: The hematopoietic reconstitution potential measured in NSG mice differed for each of the 3 UCB units tested so far. However, there was no clear impact of the pre-processing temperature on the viability of HSC, CFU and reconstitution potential, although there was a significant loss of viability in the granulocyte population.

Conclusion: Our ongoing experiments will help define the optimal conditions for UCB processing and banking.

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CD34+ CELL ENUMERATION BY FLOW CYTOMETRY: A COMPARISON OF TWO HEALTH CANADA APPROVED KITS ON AN ACCURI C6

***Marc Cloutier, Carl Simard and Sonia Néron**

Héma-Québec, R&D, Québec City, QC

Enumeration of CD34+ cells in umbilical cord blood, bone marrow or peripheral blood is an important data for critical clinical choices in stem cell transplantation therapy. Single platform cytometry assays have become the primary option for stem cell enumeration. The purpose of this study was to assess the feasibility of using commercial kits from BD and Beckman Coulter on an Accuri C6 cytometer for the enumeration of CD34+ cells in umbilical cord blood. The Stem Cell Enumeration Kit from BD and the Stemkit from Beckman Coulter were used to stain either a solution of blood doped with known quantities of CD34+ cells or a commercial stabilized leucocytes solution containing validated concentrations of CD34+ cells (Streck CD-Chex® CD34). Stained samples were analyzed on an Accuri C6 according to the ISHAGE protocole. The staining with both kits resulted in stable, sensitive, reproductible and specific results when analyzed on the Accuri C6. The regression analysis gave R-square values of 0.9997 and 0.9966 for the Beckman Coulter and BD kits, respectively. Staining of the CD-Chex® CD34 resulted in numbers of CD34+ cells inside the range given by the manufacturer. In conclusion, both the BD and Beckman Coulter kits, coupled with the cytometer Accuri C6, allow the quantification of CD34+ in ranges of concentration corresponding to the three major sources of stem cells.

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POST-REDUCTION CELL RECOVERY IN BUFFY COAT FRACTIONS OF CORD BLOOD UNITS

***Pascal Rouleau[1], Marie-Ève Rhéaume[1,3], Diane Fournier[2], Marie-Claire Chevrier[2] and Renée Bazin[1,3]**

Héma-Québec, [1]Innovation R&D and [2]Reference and Stem Cell Laboratory, Montréal, QC; [3]Laval University, Québec, QC

Background: The Héma-Québec Public Cord Blood Bank operates under guidelines established by regulatory agencies. The procedure for banking requires that each umbillical cord blood (UCB) unit is stored frozen within 48h of collection. Recently, we showed that the total nucleated cell (TNC) recovery in buffy coat (BC) prepared by volume reduction of whole UCB greatly varied between units processed shortly after collection compared to units processed at later times. Since the TNC count is an important criterium for clinical use of banked units, the present study was undertaken to better characterize the differential recovery of TNC in BC as a function of time after collection.

Method: UCB units were stored at RT for defined periods of time before processing by centrifugation at 3500g for 10 min at RT and volume reduction to 21 +/- 2 ml using the Optipress. Group 1 consisted of 16 UCB units processed within 10 h of collection (mean of 7h) and group 2 contained 10 UCB units processed from 10 up to 48h (mean of 29h) after collection. The flow cytometry ISHAGE protocol was used to establish TNC (CD45), stem cell (CD34), T cell (CD3), B cell (CD19) and granulocyte (CD15) counts as well as viability (7-AAD) in all BC and discarded red blood cell (RBC) fractions.

Results: Recovery of TNC in BC from Group 1 was 75%. However, virtually all stem cells and T cells and about 60% of B cells and granulocytes were present in the BC fractions, the remainder of B cells and granulocytes being found in RBC fractions. In contrast, TNC recovery in BC was more than 90% in Group 2; all stem cells and T cells, 85% of B cells and 80% of granulocytes were in the BC fractions, while RBC fractions contained 15% and 20% of B cells and granulocytes respectively.

Conclusion: Post-reduction recovery of stem cells and T cells in BC is excellent and is not influenced by the duration of the hold period before processing. The reduced TNC recovery in UCB units processed shortly after collection is explained by the differential distribution of B cells and granulocytes in BC and discarded RBC fraction.

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VALIDATION OF STERILITY TESTING OF CORD BLOOD AT CANADIAN BLOOD SERVICES: CHALLENGES AND RESULTS

***Sandra Ramirez-Arcos, Yuntong Kou, Heather Perkins, Lin Yang, Michael denAdmirant, Mike Halpenny and Heidi Elmoazzen**

Canadian Blood Services

Background and purpose: Canadian and International Standards mandate sterility testing for cord blood (CB) products to prevent the transmission of microbial infections to transplant recipients. This study was aimed at validating the automated BacT/ALERT 3D culture system for microbial detection in CB units processed at Canadian Blood Services.

Methods: The validation was developed in two phases. Phase 1) Fresh CB units were tested for sterility by inoculation of 8-10ml of CB into aerobic and anaerobic BacT/ ALERT culture bottles. The units were then spiked with -100 CFU/ml of the aerobic bacteria Pseudomonas aeruginosa, Klebsiella pneumoniae, Staphylococcus aureus and Staphylococcus epidermidis, the anaerobe Bacteroides fragilis and the yeast Candida albicans. CB units were processed and the byproduct plasma fraction was stored at room temperature for 24 h. Following storage, 8-10ml of plasma was inoculated into BacT/ALERT culture bottles. Phase 2) A mix of post-processing byproducts (4 ml RBCs and 4 ml plasma) was inoculated into BacT/ALERT culture bottles. Presence of the inoculated organism was confirmed in the positive cultures.

Results: During Phase 1, lack of bacterial growth was observed in units obtained from mothers that were under antibiotic treatment at the time of CB collection. Thus, antibiotic- and antifungal-free CB units were used for the remaining validation test cases. Although all bacteria were detected in plasma, C. albicans was not always captured in this fraction. In Phase 2, all organisms were detected in the mix of byproducts plasma and RBCs.

Conclusion: The Validation of CB units was successfully completed at Canadian Blood Services. The lack of growth of C. albicans in the plasma fraction was likely due to its preferential segregation to cellular fractions. The mix of byproducts plasma and RBCs is an appropriate sample for sterility testing of CB. The clinical significance of the bactericidal or bacteriostatic effect of antibiotics present in CB merits further investigation.

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CORD BLOOD STERILITY TESTING: IN SEARCH OF A WINNING FORMULA

***Marie Joëlle de Grandmont, Marie-Pierre Cayer, Mélissa Girard, Diane Fournier and Louis Thibault**

Héma-Québec, R&D, Québec City, QC; Héma-Québec, Reference and Stem Cell Laboratory, Montréal, QC

Purpose: Currently, no standard methods exist to assess cord blood (CB) for microbial contamination. Recently, we showed that about 30% of processed CB contain antibiotics which could potentially interfer with sterility test results. In addition, operational logistics of CB banks do not always allow immediate sterility testing and aliquots may sometimes wait up to 72 hours. In this work, we compared 5 different approaches to prepare CB samples for sterility testing and studied the impact of sample holding time before inoculation of culture bottles and analysis with the BacT/ALERT 3D system.

Methods: CB units (n=3) were inoculated with Klebsiella pneumoniae, Staphylococcus epidermidis, or Streptococcus agalactiae at 10 colony forming units (CFUs)/mL. Following volume reduction, the stem cell concentrate (final product) was diluted in RPMI 1640, Thioglycolate, or plasma. Two additional samples were tested: undiluted final product and plasma. Each sample were hold 24 or 72 hours at room temperature before inoculation to BacT/ALERT 3D culture bottles.

Results: RPMI 1640 and Thioglycolate media allowed bacterial detection in 89% of CB samples following the 24-hour hold storage. Thioglycolate medium seems to greater promote bacterial survival and growth during the 72-hour hold before testing. Almost half of the contaminated plasma samples (47.2%) failed to be detected by the BacT/ALERT 3D system regardless of the holding time or the bacterial strain used.

Conclusions: Although the use of RPMI medium seems to be an adequate approach to dilute CB final product for microbial detection, the sterility of samples should be performed within 24 hours following processing. The amount of plasma in test CB samples should be minimized as it seems to inhibit bacterial survival, triggering the possibility of false negative results. The choice of an effective method to prepare samples for sterility tests is crucial to obtain reliable results and safe products.

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ANTIMICROBIAL ACTIVITY OF BLOOD COMPONENTS

***Mariam Taha, Yuntong Kou, Heather Perkins and Sandra Ramirez-Arcos**

Canadian Blood Services

Background and purpose: Bacterial contamination of blood products used for transfusion is the major source of septic transfusion reactions despite the antimicrobial activities of blood components such as plasma proteins, white blood cells and platelets. This indicates that some bacteria are resistant to killing by these components. This study was aimed at evaluating bacterial susceptibility to different blood components.

Methods: Twenty-one bacteria (blood products contaminants and commercial isolates) were screened for their susceptibility to the bactericidal activity of soluble factors of different blood components using a diffusion sensitivity assay on blood agar. Blood components tested included four buffy coat fractions and their corresponding plasma units, and 11 platelet concentrates (PCs). Ten-pl drops of the blood components were placed on bacterial cultures and after incubation under ideal growth conditions, clear zones of inhibition surrounding the blood components were measured. Eight bacteria were selected and further tested for their ability to grow in the blood components in 96-well plates, where not only the soluble factors but also the cellular fractions were present.

Results: Bacteria displayed different degrees of sensitivity to different blood components. Commercial isolates of Serratia marcescens and Yersinia enterocolitica showed the greatest susceptibility while clinical isolates of Serratia liquefaciens and S. marcescens resisted elimination by all components. Some bacteria reacted differently to different units of the same product such as Streptococcus agalactiae which was inhibited by seven out of the 11 PCs tested in the diffusion sensitivity assay.

Conclusions: Our study demonstrates that the antimicrobial action of blood components varies against different bacterial species with clinical isolates showing greater resistance. Furthermore, a number of units of the same product eradicated more bacteria than ethers indicating donor variability. These data merit further investigation of the blood factors involved in antimicrobial activity that can be used to increase the safety of transfusion patients.

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TISSUE, JUST ANOTHER PRODUCT IN THE SUNNYBROOK BANK

Alison Halliday, Raj Visvalingam, *Lisa Merkley, Connie Colavecchia, Yulia Lin and Jeannie Callum

Sunnybrook Health Sciences Centre, Toronto, ON

Purpose: The ultimate goal of any transfusion service is to ensure patient safety regardless of the products being requested from the bank. This report will demonstrate how the SHSC Blood Bank successfully applied the management principles of blood products to human and animal derived tissue products within the transfusion arena, ensuring recipient safety.

Method: Existing national and international standards and regulations were applied within a quality system framework defining the tissue management processes. These processes were further defined through inter-professional collaboration with tissue user groups within the hospital (OR executive, Orthopedic, Cardiovascular and Plastic Surgery). Tissue inventories were determined, supplier qualifications were developed, ordering practices were established and receipt, inspection, storage (within appropriately monitored devices with continuing monitoring charts and alarms), tracking (within the existing Blood Bank Lab information system) and distribution policies, processes and procedures (utilizing the same issuing procedures as for blood products) and surveillance (adverse event reporting) were successfully implemented. Very little effort was required to cross train both Technical and Medical Blood Bank staff in the management of tissue products as a result of being able to apply the same blood management principles.

Results: Between January 1, 2000 and December 31, 2013, 32 tissue suppliers have been qualified and over 6000 allografts and 1400 manufactured tissue products have been issued from the Sunnybrook Blood and Tissue Bank. Despite the volume of tissue released, no adverse tissue events have been reported. As a result of an electronic tracking process any recalled tissue has been successfully accounted for in a timely manner with no patient harm or disease transmission identified.

Conclusion: The principles of blood management can easily be translated into the management of tissue products within a centralized handling and distribution model with minor impact to Blood Bank operations and major impact to improving recipient safety.

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IMPLEMENTATION OF A CONFIGURABLE LABORATORY FOR USE IN CELLULAR PROCESS DEVELOPMENT AND MANUFACTURING

***Roy, A., Boyer L. and Blais, Y.**

Direction de la production cellulaire, Laboratoire de Recherche et Développement, Héma-Québec, Québec, QC

Background aims: Héma-Québec took a new strategie direction in 2012 which aims to become a leader in cell therapy. A new process development and manufacturing lab had been implemented to transfer and optimize cellular processes and analytical methods from future Héma-Québec collaborators. Regulatory requirements for the manufacturing of cell products for clinical investigation and commercialization require a significant level of automation and record-keeping, starting early in process development and manufacturing. Central to record-keeping is the management of facilities, raw materials, processes, and assays.

Methods: To support these requirements, we evaluated several laboratory equipments, including their cost, flexibility, regulatory compliance to "GMP" requirements and ability to integrate to the newly created "Direction de la production cellulaire" . We selected the Xvivo and Quantum bioreactor systems, as they were key equipments for our laboratory in support of pre-clinical and clinical cell-production activities. We reported on the design and utilization of these systems suitable for future manufacturing operations. We established raw material and equipment listing, technical specifications and CoGs to lead to a successful transfer of technology of manufacturing processes and quality control methods from current partners.

Results: Transfer of technology and production support was initiated. We are currently working on the transfer of two products: an autologous cell therapy product and a feeder cells master cell bank.

Conclusions: Overall the new implemented lab has served to support the compliance and production requirements of process development and potential production for phase 1/11 clinical trial activities for our laboratory and can be easily customized and modified to meet the needs of future partners.

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ACQUISITION OF EQUIPMENTS FOR CONTROLLED CELL MANUFACTURING PROCESS: A STEP TO C LA VIE

***Boyer, L. Roy, A. and Blais, Y.**

Direction de la production cellulaire, Laboratoire de Recherche et Développement, Héma-Québec, Québec, QC

The Héma-Québec R&D department had recently acquired new equipments for cell processes under controlled environmental conditions to develop, optimize and transfer cell therapy processes in a future production site. Parameters control improves process traceability, reproducibility, safety and provides standardization. The current platform comprises two Quantum bioreactors (TerumoBCT) for adherent cells expansion and one isolator Xvivo (Biospherix) which is a modular closed clean room system. The Quantum bioreactor is a closed system for expansion of adherent cells. It comprises a single use hollow fiber device of 21000 cm2 surface area that automates and closes the cell culture process, reducing labor intensive tasks such as manual cell culture feeding and harvesting. This bioreactor was evaluated using human bone marrow MSC and allowed generation of approx. 300 millions cells. ISCT MSC quality attributes such as plastic adherence, phenotype and capacity to differentiate in three specific lineage were successfully evaluated. These bioreactors are currently used for scale up production of MSC from different sources. As an alternative to clean room facilities, Héma-Québec R&D department had also acquired an isolator. The modular isolator Xvivo is a classA (class 100-IS05) closed system where parameters can be controlled , monitored in real time and in a continuous mode. Our customized unit is composed of closed chambers for incubators, centrifuge, microscope and decontamination pass boxes. The environmental parameters such as temperature , pressure, humidity, particles size, volatile organic compounds and gaz (oxygen, carbon dioxide , nitrogen) can be programmed and tracked independently in each closed chamber. The entire process can be realized in a closed and controlled area similar to a clean room facility without the associated cost. The acquisition of such equipments will allow Héma-Québec to acquire an expertise in process development and optimization, cell manufacturing processes, and lead to collaborations for transfer of technology and manufacturing to the future C·LAVIE site.

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ENHANCED EXPANSION AND IMMUNOMODULATORY CAPACITIES FOR XENO-FREE-CULTURED ADIPOSE-DERIVED STROMAL/STEM CELLS

***Mathieu Drouin[1], *Nellie Dumont[1], Tony Tremblay[1], Lionel Loubaki[1], Renée Bazin[1], Julie Fradette[2] and Josée Laganière[1]**

[1]Héma-Québec, R&D Innovation, Québec, QC; [2]Centre de recherche du CHU de Québec, Axe Médecine Régénératrice et Département de Chirurgie, Université Laval, Québec, QC

Adipose-derived stromal/stem cells (ASC) offer an attractive alternative source of material for a number of cellular-based therapies, in particular for adverse graft-versus-host disease (GVHD) reactions. In this study, we first tested various culture conditions for AD-ASCs isolated from two different donors, including commercial xeno-free medium (Lonza) and matrices (fibronectin and collagen from Corning), and compiled their doubling time and total expansion potential. We observed an increase in cell proliferation for cells cultured under the commercial xeno-free medium compared to 10%-serum-containing medium. In addition, the ASC maintained expression of their characteristic cell surface markers, albeit displaying a lower expression level of CD105 in cells cultured in xeno-free conditions. Preliminary results suggest that the ASCs possess a greater immunomodulatory potential than the bone-marrow- and umbilical cord-derived MSCs, based on their capacity to suppress T cell proliferation in vitro. These results suggest that the xeno-free conditions tested in this study allow us to foresee a potential for larger-scale production of ASCs and hence ease the translation to the clinic. In addition, the great immunomodulatory potential of ASCs suggests that this cell source could be considered for the treatment of GVHD and possibly of other immune or inflammatory disorders.

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EVALUATION OF THREE METHODS FOR THE EXTRACTION OF MESENCHYMAL STEM CELLS FROM UMBILICAL CORDS

***Isabelle Paré, Lionel Loubaki, Pascal Rouleau, Hélène Émond, Nellie Dumont and Renée Bazin**

Innovation, R&D, Héma-Québec, Québec, QC

Background: Mesenchymal stem cells (MSC) are multipotent stromal cells that can differentiate into various cell types including osteoblasts, adipocytes and chondrocytes, making these cells very attractive for regenerative medicine. In addition, MSC show therapeutic effects in the treatment of acute graft vs host disease. MSC are found in many different tissues, including bone marrow and umbilical cord. However, recovery of bone marrow cells requires invasive procedures while umbilical cords are more readily available. The purpose of this work was to evaluate three methods for the extraction of MSC from umbilical cords.

Methods: Umbilical cords were obtained after informed consent has been obtained from the participating mothers. The methods evaluated were derived from protocols found in the literature and consisted in passive explantation, enzymatic digestion and extraction of vein and arteries followed by passive explantation or enzymatic digestion. All methods were tested using different sections of the same umbilical cord. A total of 9 umbilical cords were tested. The MSC extracted were cultured and their phenotype and differentiation, proliferation and immunosuppressive properties were evaluated at different intervals during the culture.

Results: Our results show that passive explantation, using either whole cord sections or vein and arteries is a reliable method for obtaining MSC but requires more time before enough cells are recovered to initiate cell expansion, compared to enzymatic digestion of the tissue. On the other hand, enzymatic digestion was less reliable since we failed to obtain MSC with a good proliferative capacity in more than half of the experiments. In our hands, there was no clear advantage to start with vein and arteries instead of whole cord sections.

Conclusion: Extraction of MSC from umbilical cords is readily achieved by passive explantation. The MSC exhibit excellent proliferative capacity, retain their ability to differentiate into various cell lineages and exhibit immunosuppressive properties in vitro.

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ASSESSMENT OF THE IMMUNOSUPPRESSIVE POTENTIAL OF MESENCHYMAL STEM CELLS BY FLOW CYTOMETRY

***Tony Tremblay, Lionel Loubaki and Renée Bazin**

Héma-Québec, R&D Innovation, Québec, QC

Background: The introduction of cellular therapies in the modern medicine has led to new approaches for the treatment of many diseases. Mesenchymal stem cells (MSC) have recently raised much interest due to their ability to exert immunosuppressive effects in the treatment of patients suffering from graft-versus-host disease (GvHD). MSCs can be obtained from several sources, including bone marrow, adipose tissue and neonatal tissues such as the umbilical cord and Wharton's jelly. The data available to date indicate that MSCs are safe and immunologically well-tolerated, but there is a need for a reliable method to establish and compare the immunosuppressive properties of MSCs obtained from these various sources. The aim of this work was to develop a cytometry-based assay to measure the effect of MSCs on activated peripheral blood mononuclear cells (PBMC).

Methods: MSCs were obtained from umbilical cords (UC-MSC) or bone marrow mononuclear cells (BM-MSC), cultured and stored frozen until use. Human PBMC were obtained from healthy volunteers after informed consent and activated with monoclonal anti-CD3/anti-CD28 antibodies in the presence or not of different MSC to PBMC ratios. After 96 hours, the extent of T cell proliferation was evaluated by establishing the number of viable T cells in the different culture conditions, using flow cytometry.

Results: Addition of anti-CD3/CD28 to PBMC resulted in a significant expansion (5-fold) of the T cell population, as evaluated following labeling with anti-CD3 APC (T cells) and 7-AAD (viability). In the presence of MSCs, T cell proliferation was significantly reduced. The inhibition was > 95% for MSCs from both sources at the 1:10 ratio. The use of the 1:20 ratio permitted to show differences in the inhibitory potential of BM-MSC and UC-MSC, the latter being more immunosuppressive.

Conclusion: We have set up a simple but very reliable assay to establish and compare the immunosuppressive potential of MSCs from various origins. In future work, the immunosuppressive potentiel of UC-MSC and BM-MSC will also be tested in an animal model of GvHD.

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