Monday 15 October **0830-1000**
HSANZ Free Communications 1: Benign Haematology **Arena 1B**

O01 0830

Observational Study of Iron Overload as Assessed by Magnetic Resonance Imaging (MRI) in an Adult Population of Transfusion Dependent Patients with β Thalassaemia: Significant Association Between Low Cardiac T2* <10ms and the Occurrence of Cardiac Events

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Aim
To assess the significance of cardiac iron overload as assessed by MRI T2* in a regional adult Australian population of patients with transfusion dependent β thalassaemia. This is relevant as such patients usually succumb from cardiac events.

Method
Ethics committee approved observational study of cardiac iron overload as assessed by MRI cardiac T2* relaxometry in 30 transfusion dependent β thalassaemia patients: Evaluation using correlation coefficients, unpaired t test, Fisher exact test and log rank time to event analysis with cardiac events (congestive heart failure, arrythmia, cardiac death), left ventricular ejection fraction (LVEF), serum ferritin (SF) and MRI assessed liver iron concentration (LIC). 29/30 MRI measurements were performed between October 2003 and September 2005.

Results
11/30 patients (37%) had cardiac T2* <10mS, 8/30 (27%) in range 10-20mS and 11/30 (37%) >20mS. There was no significant correlation between T2* values and values for SF(P=0.26), LIC(P=0.53), age(P=0.12) and LVEF (P=0.12). Baseline mean (CI) LVEF values were 49% (2.8) in patients with T2* <10mS and 57% (2.3) in patients with T2*>10mS (P=0.01). Very low T2* values <10mS were strongly associated with the occurrence of cardiac events: occurring in 5/11 in patients with T2* <10mS and in 0/19 in patients with T2* >10mS (P=0.003 Fisher exact test; P=0.001 log rank Kaplan-Meier time to event analysis). There was no significant association between T2* <10mS or cardiac events and traditional measures of iron overload such as SF levels >2500mcg/l (P=0.1), LIC (evaluated at thresholds of >7 or >15mg/g dry weight) or with age above or below median value of 32 years.

Conclusion
Very low cardiac T2* values <10mS are commonly seen in adult populations of β thalassaemia and are significantly associated with risk of cardiac events. This investigation permits the application of individually targeted iron chelation strategies which are more effective in removing cardiac iron.

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O02 0845

Individually Guided Iron Chelation Therapy in a Transfusion Dependent Adult Population With β Thalassaemia Following Assessment of Iron Overload by Magnetic Resonance Imaging (MRI)

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Aim
To individually target iron chelation therapy according to organ specific iron overload data assessed by MRI techniques and newly available treatment options for iron chelator choice.

Method
Audit of the use of organ specific iron overload data, assessed by Ferriscan ® liver iron quantification [1], and myocardial T2* relaxometry [2], to guide individual iron chelator treatment options. Use of paired t test, Fisher exact test and correlation coefficient.

Results
At the time of initial analysis (n=30) 43% of liver iron concentration (LIC) values were < 3 (mg/g dry weight); 17% in range 3 – 7; 20% in range 7 - 15 and 20% >15 mg/g/dry weight, representing ranges suggested to be significant for management decisions [3]. There was a significant correlation between LIC and serum ferritin (P=0.001) but no significant correlation or association of LIC with current or subsequent cardiac events, cardiac T2* or left ventricular function. Documentation of very high LIC, cardiac events and later low cardiac T2* (refer other abstract) prompted change in chelation practice from historical s.c. desferrioxamine (DFO) to alternatives such as i.v. DFO or combination DFO and deferiprone. Examples of individual patients showing improvement in various iron overload parameters will be shown to illustrate the principle of risk adapted individualised iron chelation therapy.

Conclusion
Recent data have confirmed the significance of organ specific iron overload data as assessed by MRI and the improved cardioprotective effects of new iron chelator treatment options [4]. This permits the use of more effective iron chelator therapy to those at greatest risk of cardiac disease.

References
1 Resonance Health Analysis Services Perth Australia www.ferriscan.com
2 Brown G, et al. Proc International Society for Magnetic Resonance in Medicine (ISMRM), 2004;1668.
3 Olivieri N, Brittenham G. Blood 1997; 89:739-761.
4 Neufeld EJ. Blood 2006:107:3436-41.

Monday 15 October **0830-1000**
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O03 0900

Clinical Outcomes and Adherence in Thalassaemia Major Patients, A Single Institution Audit

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Aim
To evaluate all transfusion-dependent thalassaemia patients attending a single institution for iron-overload complications, adherence to chelating agents, general health outcomes and complications of transfusion. Of special interest is the interaction between adherence to subcutaneous and oral chelating agents.

Method
Forty-four patients were assessed retrospectively and prospectively over a 6 year period. All patients completed a questionnaire. Pharmacy dispensing records were evaluated for desferrioxamine and deferiprone. 'Percent dispensed' was derived by dividing the amount of chelating agent dispensed over the amount prescribed. Clinical review, bone mineral densitometry, gated heart pool scan, blood bank records and routine surveillance investigations were included. Serial ferritin measurements were collated.

Results
This cohort demonstrates equivalent or improved health outcomes compared to other registries, however, there continues to be significant morbidity. The prevalence of diabetes mellitus is 18%, hypothyroidism 16%, hypogonadism 32% and cardiomyopathy 5%. Osteopenia/osteoporosis is present in 83%. Serological evidence of exposure to Hepatitis C and Hepatitis B occurred in 41% and 14% of patients respectively. Twenty-three percent of patients have active Hepatitis C. Predictors of complications include poor adherence, ferritin >2000 µg/L over the preceding six years, increasing years of transfusion and increasing age.

There was a wide range of adherence to desferrioxamine (0-100% of prescribed dose, mean 49%, median 46%), and deferiprone (29-214%). Poor adherence, particularly with desferrioxamine, correlated with failure to attend for regular clinical review and these patients were more likely to overstate their usage in the questionnaire. Of note, patients in whom concomitant deferiprone was prescribed had a significantly lower desferrioxamine dispensing rate ($p<0.006$). Collection of desferrioxamine from pharmacy was reduced in the year following commencement of deferiprone.

Conclusion
Despite improvements in management, transfusion-dependent thalassaemia major patients continue to have endocrinopathies and cardiac disease related to iron-overload. Osteoporosis affects the majority of patients, especially those who are well chelated. Adherence to therapy is a major determinant of long-term complications, and an important area to maximize. The interaction between desferrioxamine and deferiprone prescription is complex and oral therapy tends to reduce compliance with subcutaneous therapy.

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O04 0915

The Therapeutic Penetrance of HFE Haemochromatosis Genotypes: What is and what is not Haemochromatosis

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Aim
Haemochromatosis (HH) is a very common inherited disorder of iron overload. The major problem preventing widespread genetic screening for HH is the variable disease penetrance seen in C282Y homozygotes. Not everyone who is a C282Y homozygote develops HH (incomplete penetrance). The aims of this study was to establish biochemical and "*therapeutic"* penetrance of the C282Y homozygous genotype (HFE) that is responsible for about 80% of HH cases in Western European descents. Compound heterozygous (C282Y/H63D) were also studied and potential genetic modifiers of the penetrance were considered in this group.

Methods
We have undertaken clinical (symptoms and questionnaire), biochemical (transferrin saturation and ferritin levels) and genetic studies (genotyping, DNA sequencing) between 1997 and 2006 in 810 C282Y homozygous and 175 compound heterozygous (C282Y/H63D) unrelated individuals previously undiagnosed with haemochromatosis. Biochemical penetrance was established according to elevated iron indices (Tsat % > 55% ; ferritin > 320 □g:L^{-1} in adult males and Tsat % > 50 %; ferritin >250 □gL^{-1} in premenopausal women). Removal of a minimum of 5 gm iron was considered to be an index of full "therapeutic" penetrance.

Results
Biochemical penetrance, was much higher than the clinical penetrance. Indeed, most C282Y homozygotes displayed a common biochemical phenotype, namely an elevated Tsat level which can be found with an increased serum ferritin level in up to 77 % of men (173) and 56% of women (51). The therapeutic penetrance was established at 0.25 in our population of C282Y homozygous individuals (810) with a significant difference between males (0.36) and females (0.14). Among individuals with the C282Y/H63D (n=175) biochemical penetrance was 20 % (n=35). Eight percent of C282Y/H63D individuals presented with classical HH phenotype, and half of them were found to have either a supplementary mutation in HFE or a mutation in another iron regulatory gene (digenic inheritance).

Conclusions
In the EU it is calculated that there are about 1.2 million people homozygous for the C282Y mutation, which would be expected to result in more than 200 000 individuals needing treatment according to our definition of the penetrance, with implications for public health.

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O05 0930

Is Haemoglobin a Good Indicator of Iron Status in Women of Reproductive Age in North-Western Vietnam?

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Aim
Iron deficiency anaemia poses a risk to women of reproductive age (WRA) in developing countries. The World Health Organization (WHO) recommends epidemiologic assessment of haemoglobin (Hb) to evaluate iron supplementation programs, as resources for measurement of iron indices are often unavailable. We evaluated a) haemoglobin as a test of iron deficiency, and b) predictive indices of supplementation impact, during a weekly iron/ folate supplementation (WIFS) program in north-western Vietnam.

Method
Haemoglobin, ferritin and transferrin receptor (TfR) were evaluated, and the transferrin receptor/$\log_{10}$Ferritin Index (TfR/F Index) calculated before and after institution of weekly ferrous sulphate (60mg) and folic acid (400mcg) supplementation. Anaemia (Hb<12g/dL) and iron deficiency (Ferritin<15ng/mL) were defined using WHO guidelines. Iron deficiency was also defined by alternative criteria (TfR>2.3mcg/mL, TfR/F Index>1.8) cited in recent literature. Proportions of anaemic WRA with iron deficiency; and area under ROC curves (AUC^{ROC}) of haemoglobin to identify iron replete states were calculated. Baseline iron indices of anaemic women who achieved a 'clinical response' (CR- Hb increase ≥1.0g/dL, or resolution of anaemia) were compared to 'non response' (NR).

Results
At baseline, 131/349(37.5%) women were anaemic. Of anaemic women, 47/123 had ferritin<15ng/mL (PPV=38.2%[29.6-46.9]). AUC^{ROC} of haemoglobin to detect iron repletion was 0.70[0.63,0.77]. A TfR>2.3mcg/mL was observed in 48/126(PPV=38.1%[29.6-46.6]) anaemic WRA (AUC^{ROC} 0.731[0.661,0.801]) and 45/122(PPV=36.9%[28.3-45.5]) had a TfR/F Index>1.8 (AUC^{ROC}0.769[0.701,0.837]). Of 219 women re-evaluated after three months of WIFS, 81 (37.2%) had been anaemic at baseline; 55/77 (71.4%) of these anaemic women achieved CR. Comparing CR to NR anaemic groups, median baseline ferritin was 18ng/mL vs 33.5ng/mL, p<0.05 (Wilcoxon rank-sum) and TfR/F Index 1.32 vs 1.09, p<0.02.

Conclusions
Fewer than half of anaemic women were iron deficient using WHO criteria. Haemoglobin was of intermediate value for predicting iron status. Iron indices, particularly ferritin and Tfr-F index, identify anaemic WRA who responded to supplementation, and ideally should be measured in epidemiologic studies.

Monday 15 October **0830-1000**
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O06 0945

Characterisation of the Bone Marrow Immunofluorescence Test in Childhood Autoimmune Neutropenia

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Aim
The bone marrow immunofluorescenece test (BMIFT) [1] demonstrates autoantibodies to granulocytes and their precursors on fresh frozen bone marrow slides. It may be used to differentiate childhood autoimmune neutropenia (AIN) from other causes of childhood neutropenia, even when circulating neutrophil counts are low. We have sought to characterise the diagnostic utility of the BMIFT in childhood AIN.

Methods
All BMIFT requests for investigation of children with neutropenia between January 1998 and May 2007 were reviewed. Patients were classified as AIN or non-autoimmune causes. Baseline demographic data, results of BMIFT, granulocyte immunofluorescence testing (GIFT) and bone marrow findings were collected from clinical records and the institutional laboratory database. Two-by-two table analysis was performed to determine the sensitivity, specificity, positive (PPV) and negative predictive value (NPV) for each test.

Results
Seventy-six children had BMIFT performed for investigation of neutropenia. There were 45 patients diagnosed with AIN, 28 with non-immune neutropenia and 3 failed tests. Non-autoimmune causes included transient neutropenia (sepsis or neonatal) (n=6), drugs (n=5), chronic benign neutropenia (n=5), viral (n=4), congenital severe (n=3), cyclic (n=2), neonatal alloimmune (n=2) and aleukaemic ALL (n=1). The median age of children with AIN was 1.2 years (range 0.3 - 15.3), compared to 3.6 years (range 0.1-15.7) in the non-autoimmune group. The median neutrophil count in AIN was 0.3x10^9/L (0.9x10^9/L in non-autoimmune). BMIFT was positive in 24 of 45 patients with AIN and 0 of 28 with non-autoimmune neutropenia (sensitivity 53%, specificity 100%, PPV 100%, NPV 57%). GIFT was positive in 10 of 24 patients with AIN and 0 of 9 with non-autoimmune neutropenia (sensitivity 42%, specificity 100%, PPV 100%, NPV 39%). Bone marrow findings in AIN included maturation arrest (n=7), eosinophilia (n=3), left shift (n=7) and normal (n=28). Twelve patients had other autoimmune diatheses at diagnosis.

Conclusion
The BMIFT is a simple, highly specific test with excellent positive predictive value and thus is a clinically useful test to confirm autoimmune neutropenia in children.

Reference
1 Johnson T, Minchinton R. Investigating Severe Neutropenia With a Simple Bone Marrow Immunofluorescence Test. British Journal of Haematology 1997;99:418-21.

Monday 15 October **0830-1000**
HSANZ Free Communications 2: Cellular Immunotherapy **Meeting Room 7**

O07 0830

RNA Loaded CD34+Derived DC Generate Anti-Leukaemic CTL That Alter the Kinetics of Leukaemic Growth In Vivo

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Aim
An immunotherapeutic approach that enhances the graft-versus-leukemia effect may improve the survival of patients with Acute Lymphocytic Leukaemia (ALL) patients who relapse after transplantation. We postulate that CTL generated from total RNA loaded cord blood CD34+-derived dendritic cells (CD34+DC) can control the kinetics of leukaemic growth in a NOD-SCID mouse model of human ALL.

Methods
We electroporated CD34+DC with total RNA from a patient ALL xenograft to generate patient-specific anti-leukaemic CTL. NOD-SCID mice were inoculated with $1x10^5$ ALL xenograft cells±anti-leukaemic CTL and the kinetics of ALL growth were monitored in vivo.

Results
Anti-leukaemic CTL, consisting of CD4 (69.9±2%), CD8 (12.9±2%) and NK cells (17.6±1%), lysed 63±6% of ALL xenograft targets and 22±7% autologous MNC (p=0.04). Administration of anti-leukaemic CTL ($20x10^6$ cells per mouse) 2, 3, 4 and 5 weeks post ALL inoculation, altered the kinetics of ALL growth in vivo (p=0.00032). By 8 weeks post ALL xenograft, mice given ALL alone had 3.6% $CD45^+CD19^+$ ALL cells in the blood, while ALL-bearing mice who received CTL had ALL cells (0.66%) in the blood (p=0.05). After 11 weeks, the ultimate disease load was the same in mice who received ALL alone and those mice who received 4 doses of 20 x 10^6 CTL. Evaluation of the kinetics of engraftment of the ALL xenograft showed that cells homed rapidly to the spleen (6.9±1%) and bone marrow (1.6±1%) after 1 week but were only detectable in the blood after 6 weeks (1.8±2%). By 7 weeks, the spleen and bone marrow (>55%) were infiltrated with ALL cells while only 5.9±2% ALL cells were detectable in the blood. A timed sacrifice experiment following the fate of infused CTL showed that CTL were readily detectable in the spleen after 3 consecutive CTL infusions ($CD45^+CD19^-CD8^+$ 20%, $CD45^+CD19^-CD4^+$ 5% and $CD45^+CD19^+$ ALL cells 22%).

Conclusions
Anti-leukaemic CTL generated from RNA-loaded CD34+DC control leukaemia in the NOD-SCID mouse model. Whilst the CTL persisted in the mouse they ultimately failed to control ALL growth because the xenograft expanded so rapidly. Disease control may be further enhanced by antigen-specific restimulation of the CTL and/or refined administration in a minimal residual disease setting.

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Monday 15 October **0830-1000**
HSANZ Free Communications 2: Cellular Immunotherapy **Meeting Room 7**

O08 0845

Antibody to Activated Human Dendritic Cells Prevents Acute Graft-versus-Host Disease

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Allogeneic hematopoietic stem cell transplantation is a powerful therapy limited by graft-versus-host disease (GVHD) in which dendritic cell (DC) stimulation of donor T cells plays a major causative role. Current immunosuppressive measures to control GVHD target T cells but compromise post-transplant T cell mediated immunity in the patient, particularly to cytomegalovirus (CMV) infection.

Aim
To investigate the activated DC as a potential therapeutic target for GVHD.

Methods
We used allogeneic mixed lymphocyte culture (MLC) as an *in vitro* model of GVHD. For *in vivo* studies, we used the human T cell dependent peripheral blood mononuclear cell transplanted SCID mouse (hu-SCID) model of GVHD.

Results
Treatment of MLCs with CD83 antibody that depletes activated human DC, suppressed allogeneic T cell proliferation but preserved T cells and retained specific cytotoxic T cell responsiveness to CMV and influenza virus. Campath-1H treatment of MLCs also suppressed allo-proliferation but T cell numbers and responsiveness to viral antigens were reduced.

We found that the hu-SCID model requires human DC for GVHD and that treatment with the CD83 antibody prevented GVHD but, in contrast to Campath-1H and antithymocyte globulin (ATG) treatment, did not prevent engraftment of human T cells, including CMV specific $CD8^+$ T cells.

Conclusion
Antibodies that target activated DC are a promising new therapeutic approach to the control of GVHD.

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O09 0900

Placental Derived Mesenchymal Stromal Cells Enhance Umbilical Cord Blood Engraftment

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Umbilical cord blood transplantation (UCBT) is an alternative to unrelated stem cell transplantation in paediatric patients with haematological malignancies. In adults the limiting factor is inadequate cell dose. Co-transplantation of bone marrow (BM) mesenchymal stromal cells (MSC) has been shown to enhance engraftment of UCBT. BM MSCs are well characterised but are obtained after an invasive procedure and the MSC number decreases with age. The placenta may provide an alternative source of MSCs but placental derived MSCs are poorly characterised. Their use in UCBT warrants investigation. In this study we have characterised placental MSCs and assessed their role in UCBT in a non-obese diabetic/severely immuno-deficient (NOD/SCID) mouse model.

MSCs were obtained by enzymatic digestion of placental tissue and isolated by selective plastic adherence. Cell morphology, immunophenotype, and proliferation and differentiation potential and karyotype were studied.

NOD/SCID mice underwent UCBT following sublethal irradiation. There were four treatment groups who received either single UCBT or double UCBT in equivalent doses (5 x 10^5 CD34+ cells) either alone or with MSCs. Mice were sacrificed 7 weeks after transplantation and engraftment assessed by flow cytometry and cellular origin by STR PCR chimerism analysis.

Placental MSCs demonstrated fibroblastic morphology, immunophenotype and differentiation potential as BM MSCs.

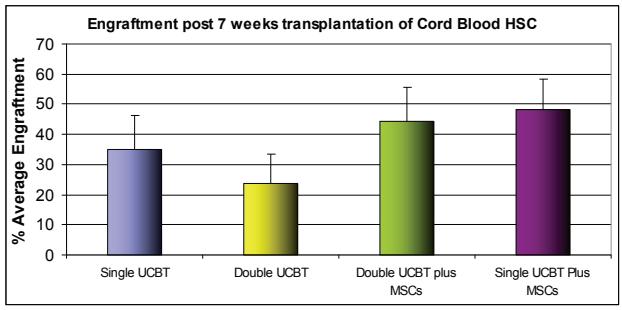

Figure .1: Haematopoietic engraftment of single and double UCBT with and without MSCs.
Mice who received MSCs along with either single or double UCBT showed increased engraftment. Transplantation of double UCBT did not improve engraftment when compared with a single UCBT of equivalent dose.

Conclusion
Placental MSCs demonstrate similar morphological, immunophenotypical and differentiion characteristics to BM MSCs.
At equivalent cell dose single and double UCBT lead to similar engraftment.
Placental MSCs improve engraftment in both settings.

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HSANZ Free Communications 2: Cellular Immunotherapy **Meeting Room 7**

O10 0915

Immunotherapy for Prophylaxis of CMV Following Allogeneic Haemopoietic Stem Cell Transplantation Using Donor Lymphocytes Stimulated with Gene Modified Dendritic Cells (DC)

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Background
Cytomegalovirus (CMV) reactivation post-allogeneic haemopoietic stem cell transplant (HSCT) continues to cause morbidity and mortality. Pharmacological therapy is limited by side effects. Adoptive transfer of *ex-vivo* generated CMV-specific T-cells has the potential to restore immunity, prevent CMV and circumvent the need for pharmacotherapy.

Methods
We have generated donor-derived CMV-specific T-cells for 16 patients using dendritic cells transduced with an adenoviral vector encoding the immunodominant pp65 protein and have infused 7 patients with this product. Previously we generated CMV-specific T-cells using DC pulsed with the HLA-A2 restricted nonapeptide NLVPMVATV (NLV) and infused the product to 9 patients. T-cells were administered prophylactically post-HSCT. Recipients were monitored for adverse reactions, CMV reactivation by polymerase chain reaction and CMV-specific immune reconstitution.

Results
Using tetramer and ELISPOT analysis we have demonstrated rapid, polyepitope, CMV pp65-specific immune reconstitution post infusion in those patients receiving adenovirus stimulated T cells. CMV reactivation by PCR has occurred in 3 patients but none have progressed to CMV disease, suggesting the functional capacity of the cells to control viral reactivation. One patient received pharmacotherapy with valaciclovir while no other patients have received anti-viral antibiotics. Of those receiving NLV-specific T-cells, 2 recipients developed CMV reactivation post-infusion but neither developed CMV disease or required pharmacotherapy. A rise in NLV-specific T-cells was demonstrated by NLV-tetramer and ELISPOT analysis in the majority of recipients. This persisted for several weeks before returning to baseline values. There have been no infusion related adverse reactions. Graft versus host disease, non-CMV infections and other adverse events did not exceed expected rates.

Conclusions
Prophylactic adoptive transfer of CMV-specific T-cells is safe, hastens CMV-specific immune reconstitution and may reduce the need for anti-CMV pharmacotherapy in allogeneic HSCT recipients. Our data indicate the potential of specific cellular therapy to control opportunistic infections in severely immunosuppressed patients post-HSCT.

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HSANZ Free Communications 2: Cellular Immunotherapy **Meeting Room 7**

O11 0930

An Adenoviral Vector Encoding the Cytomegalovirus (CMV) Matrix Protein pp65 Generates Bi-virus Specific T cells Useful for Prophylactic Immunotherapy After Allogeneic Haemopoietic Stem Cell Transplant (HSCT)

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Introduction
Since October 2006 we have conducted a phase I/II clinical trial of *ex vivo* expanded donor derived CMV specific T cells given prophylactically to HSCT recipients. Donor T cells were stimulated with monocyte derived dendritic cells transfected with Ad5F35pp65, a recombinant adenovirus encoding the entire CMV antigen pp65 that promotes presentation of a broad range of pp65 epitopes in patients of all HLA types. Quality assurance testing prior to infusion involved tests for microbial contamination, phenotype, viability and alloreactivity by ^{51}Chromium release assay.

Results and Discussion
To date, fourteen cultures have been completed. There was an 8.5-30 fold increase in total cell number (mean of 2.2 x 10^8 cells, range 1.2-5.1 x 10^8). Products consisted of T cells (median 92%) displaying an effector memory phenotype ($CD45RO^+$, $CD62L^-$) and a predominance of $CD8^+$ (14-90%) over $CD4^+$ (2.5-57%) cells. All cultures exhibited negligible alloreactivity against recipient derived targets (0-3.6% specific lysis at E:T ratio of 20:1) but strong killing of CMV targets (specific lysis against targets labeled with an overlapping peptide mix of pp65 was 43-79% at E:T 20:1). Killing was also observed against targets labeled with adenoviral antigens (2.7-12.5% lysis at E:T 20:1) demonstrating the bi-virus specificity of these cultures. In three HLA-B7 donors, T cells recognizing the pp65 epitope TPRVTGGGAM accounted for 9, 13.8 and 22.8% of all cells, corresponding to a 550-1150 fold expansion of cells recognizing this epitope. In HLA-A2 donors, T cells recognizing NLVPMVATV accounted for up to 18.2% of cells. Enrichment of cells recognizing known HLA-A24, B8 and B35 epitopes was not observed despite these cultures exhibiting strong cytotoxic activity against pp65 pulsed targets. To date, seven patients have received T cell infusions at day 30 to 101 post transplant and *in vivo* analysis demonstrates reconstitution of virus specific immunity.

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O12 0945

Expansion of Functional T cells Specific to Varicella-zoster Virus and *Aspergillus fumigatus* for Potential Use in Adoptive Immunotherapy

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Varicella-zoster virus (VZV) and *Aspergillus fumigatus* are both important opportunistic pathogens that cause major morbidity and mortality in transplant recipients. VZV is an alphaherpesvirus that establishes a latent infection in the dorsal root ganglia following an acute infection, and it can result in post herpetic neuralgia upon reactivation. *A.fumigatus* is a mold that produces airborne conidia (asexual spores) that are readily inhaled into the lungs and in immunosuppressed patients can lead to invasive aspergillosis. Previous studies have shown that lymphocytes provide a critical secondary defense against these pathogens; adoptive transfer of VZV and Aspergillus specific T cells potentially restores adaptive immune responses in transplant recipients. To generate antigen specific T cells, PBMCs isolated from normal donors were stimulated using a live attenuated VZV strain Oka vaccine (VARIVAX®, 1350PFU per vial) or heat-inactivated *A.fumigatus* conidia, re-stimulated with pulsed PBMCs after 1 week, and cultured for an additional 2 weeks in the presence of IL-2. For VZV, various concentrations of VARIVAX were tested to stimulate PBMCs, and the optimal concentration was determined to be 500PFU per 10^6 PBMCs. To date, a 10-fold expansion was observed in VARIVAX stimulated PBMCs containing a majority of $CD3^+/CD4^+$ cells (82%), with 98% of these cells expressing CD45RO. For Aspergillus, the optimal concentration was found to be one conidium per PBMC, and this generated a 6-fold expansion of PBMCs after 3 weeks of culture. 77.3% of these cells were $CD3^+/CD4^+$, with 88% expressing CD45RO. An intracellular IFN-g production assay using VZV lysate or Aspergillus conidia as stimulus was performed to test antigen specificity on expanded cultures, and 7.6% and 8.4% of CD4+ T cells were positive for IFN-□, respectively. This ongoing study demonstrates the generation of T cells specific to VZV and Aspergillus, and further studies will determine if these cells could be used in protecting against VZV and Aspergillus manifestations in transplant patients.

Monday 15 October **0830-1000**
HSANZ Free Communications 3: Leukaemia **Meeting Room 8**

O13 0830

Combined Valproic Acid and Low Dose Cytarabine is a Novel Low-intensity Regimen with Activity in Elderly Acute Myeloid Leukaemia

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Aim
Elderly patients with acute myeloid leukaemia (AML), and those with relapsed/ refractory disease have particularly clinical outcomes. Valproic acid (VA) is a histone deacetylase inhibitor with preclinical efficacy in a variety of tumours. We examined the in vitro activity and mechanism of combined VA and cytarabine and used these results to develop a novel low-intensity regimen for elderly patients with AML.

Methods
In vitro experiments: AML cell line models were used to develop and validate assays to measure apoptosis, cytostatic effects, differentiation and acetyl histone-H3 induction. Primary AML cells were isolated from blood and bone marrow from patients with AML and cultured for 24 hours. Cells were treated with cytarabine (0, 0.5 or 5μM) with or without VA for a further 24 hours and analysed. Clinical trial: Elderly patients with AML, unsuitable for intensive therapy, were treated with oral VA (200mg tds, titrated) and subcutaneous cytarabine (10mg/m^2 d1-14) in 28 day cycles. Patients were monitored for response, VA levels and toxicity. Samples were collected at baseline, d8 and d28 for in vivo histone acetylation analysis. Patients were offered up to 3 cycles of treatment.

Results
In vitro experiments: Cell lines and 11 patient samples were analysed. VA and combination therapy enhanced apoptosis (mean baseline 40%, VA 53% and combination 67%, p=0.01) and inhibited cell proliferation with VA alone (p<0.01) but not with combination (p=0.28). Acetyl histone-H3 induction was seen in some samples. VA was more effective in patients with favourable or standard risk cytogenetics (median difference 15% vs 5%)

Clinical trial: Thirteen patients were enrolled and 12 have completed treatment, with median age 72 (range 67-84). Twelve patients had ≥1 adverse prognostic feature in addition to age >60 (4 relapsed/refractory, 4 poor risk cytogenetics, 3 prior MDS and 6 performance status ≥2). The median survival was 94 days. Five patients have had a favourable response to therapy; 3 with haematological improvements and 4 with reduction in blast percentage. Four favourable responses were in standard cytogenetic risk patients. There were no complete remissions. There were 3 deaths during treatment, 2 from progressive disease and one from intracranial haemorrhage. There was no other serious non-haematological toxicity.

Conclusions
VA and low dose cytarabine is a rational combination therapy based on in vitro efficacy and appears to be a well tolerated, novel low-intensity regimen with clinical activity in elderly poor prognosis AML.

Monday 15 October **0830-1000**
HSANZ Free Communications 3: Leukaemia **Meeting Room 8**

O14 0845

An Induction Mortality Score in Elderly Patients with Newly Diagnosed Acute Myeloid Leukemia Undergoing Intensive Chemotherapy

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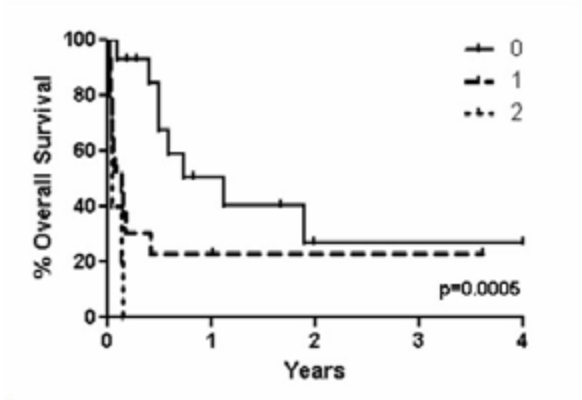
Standard therapy for "fit" elderly patients with newly diagnosed acute myeloid leukemia (AML) includes intensive anthracycline and cytarabine-based combination chemotherapy. Patients with poor performance status and extreme age are usually considered "unfit" for intensive chemotherapy due to excessive risk of induction toxicity. Despite exclusion of such patients, induction mortality in elderly AML patients treated with intensive chemotherapy remains significant. Better predictors of induction mortality in elderly patients would be extremely useful, allowing vulnerable patients to avoid the risk of early death and receive less toxic therapies. The aim of the present study was to identify pre-treatment clinical predictors of early (<42 days) death in newly diagnosed elderly non-M3 AML patients considered "fit" for intensive chemotherapy (age 56-75 years, performance status 0-2). Parameters assessed at baseline included patient specific factors: age, sex, body mass index, hemoglobin, inflammatory markers (fever, CRP, albumin), hemostasis (APTT, INR, TCT, fibrinogen, D-Dimer, platelet count), and the presence of pulmonary infiltrates, liver dysfunction (ALT, bilirubin, ALP, GGT), electrolyte disturbance (sodium, potassium, bicarbonate, calcium) or renal impairment. Disease-specific factors studied included cytogenetics, associated myelodysplasia, tumor burden (total WCC, LDH, % bone marrow blasts) and type of anthracycline received (daunorubicin or amsacrine). The median overall survival of the 36 patients included in the study was 5 months. Induction-related mortality was 27%. Univariate analysis for predictors of early death identified WCC >50 x 10^9/L, obesity (BMI >30), renal impairment (GFR <50 ml/min), INR >1.4 and albumin <33 g/L as prognostically relevant ($p \leq 0.05$). After multivariate analysis, only high leukocyte count (hazard ratio 2.3; p=0.02) and obesity (HR 2.5; p=0.02) retained statistical significance. An AML induction mortality score (AIMS) was developed based on the presence of WCC >50 and BMI >30 at diagnosis. "Fit elderly" AML patients with 0,1 or 2 risk factors had induction mortality rates of 0%, 40% and 80%, and median overall survival of 13, 2 and <1 month, respectively (Figure 1). In conclusion, the AML induction mortality score is a simple method for identifying elderly patients at high risk for early death after intensive chemotherapy and may help identify patients more suited to less toxic therapies.

Figure 1



Monday 15 October **0830-1000**
HSANZ Free Communications 3: Leukaemia **Meeting Room 8**

O15 0900

Treatment of Secondary Acute Leukaemia: The 2001-2006 Royal Brisbane Hospital Experience

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Aims
To review the outcomes of secondary acute myeloid leukaemia (AML) treated at our institution.

Method
All patients who received any chemotherapy for a diagnosis of secondary AML between 2001-2006 were retrospectively reviewed. Patients were identified from our departmental leukaemia database.

Results
In total, 27 patients were identified, including 18 (67%) males and 9 (33%) females. Fifteen patients (56%) were aged ≥60yrs. Predisposing disorder prior to AML diagnosis was MDS in 12 cases (44%), MPD in 11 (41%, including 4 patients [15%] with CML in myeloid blast crisis), prior chemotherapy for solid organ malignancy in 3 (11%) and congenital neutropenia in 1 (4%). Adverse karyotypic findings were found in half of 24 patients with diagnostic cytogenetic data available, including a complex karyotype (≥3 abnormalities) in 9 cases (38%) and chromosome 7 and 5 deletions in an additional 2 and 1 patients respectively (12%). Induction chemotherapy included standard "7+3" in 10 cases (37%), HiDAC-based regimens in 14 (52%) and hydroxyurea / oral melphalan in 3 (11%). Overall 11/27 (41%) patients achieved remission, including 9/12 patients <60yrs and 2/15 patients ≥60yrs ($p<0.002$). Chemotherapy related mortality (CRM) was also greater in patients ≥60yrs (53% vs 16% respectively, $p=0.1$). Four patients <60yrs received a subsequent allogeneic stem-cell transplant (SCT). Only 2/12 patients (16%) aged <60yrs remain alive (at follow-up 31 and 65mths respectively), including 1/4 patients who received an allogeneic SCT. Median survival for patients <60yrs was 11mths (range 0-65mths). All patients ≥60yrs have died, with only 2/15 (16%) surviving >12mths. Interestingly, both elderly patients surviving >12mths were refractory to induction chemotherapy. Median survival for patients >60yrs was 2mths (range 0-17mths).

Conclusions
Outcomes of conventional therapy for secondary AML are dismal, especially in elderly (≥60yrs) patients. Alternative approaches to treatment of secondary AML are urgently required.

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Monday 15 October **0830-1000**
HSANZ Free Communications 3: Leukaemia **Meeting Room 8**

O16 0915

Use of Interferon Therapy as Induction and Maintenance Therapy for Patients with Hairy Cell Leukemia (HCL)

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Aim
In this prospective study, role of use of interferon therapy as a single agent in induction and maintenance therapy is examined. Recent trend of use of purine analogs is not without failure (up to 36% many require further therapy) and possible association with therapy related myelodysplasia or acute myeloid leukaemia (AML), which prompted this review.

Method
Patients with a confirmed diagnosis of HCL were offered to commence on interferon therapy at 3 million units three times a week. Once haematologic response was achieved including normalisation of full blood count and splenic size, maintenance therapy at 3 mu once a week was commenced. Follow up bonemarrow biopsy was performed if any concerns or cytopenias and abdominal ultrasound or CT scans were performed to assess splenic size.

Results
Ten patients with the diagnosis of HCL were enrolled to commence on the therapy. One patient withdrew and chose not to undertake any therapy and another chose to trial 2-chlorodeoxyadenosine (2-Cda). Eight evaluable patients were followed up on this therapy. There were 3 female and 5 male patients with a mean age of 50.5 years (range: 41-64). These patients were followed up with regular FBC, ultrasound or CT scans and bone marrow biopsies when appropriate. Follow up period ranged between 3 and 15 years with a mean of 9 years. 7 patients achieved complete remission and 1 patient partial remission. These patients tolerated the therapy reasonably well.

Conclusion
HCL is a rare indolent chronic B cell lymphoproliferative disorder. There are various therapy options available. Immunosupressive effect of purine analogs increases risk of infections and there are case reports of associated myelodysplasia and AML as well as relapse requiring further therapy in up to 36% of patients.

This study has shown that use of interferon as an induction agent followed by low dose maintenance therapy is useful in achieving and maintaining CR. In this treatment population 7/8 (90%) achieved and maintained remission with a mean follow up of 9 years. In patients with HCL, interferon therapy should be considered as a safe therapy option. It may have a role in maintenance therapy in patients who fail the achieve CR with other therapies.

Monday 15 October **0830-1000**
HSANZ Free Communications 3: Leukaemia **Meeting Room 8**

O17 0930

The Detection of Nucleophosmin-1 (NPM) and fms-like Tyrosine-kinase-3 (FLT3) Mutations in Normal Karyotype Acute Myeloid Leukaemia (NK-AML) Patients Using High Resolution Melting (HRM) Analysis

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Background
Molecular characterisation in NK-AML is an essential requirement for determining prognosis. NPM1 is a nucleolar phosphoprotein involved in cell proliferation and growth-suppression: 45%-60% of NK-AML patients have an NPM1 mutation and a favourable clinical outcome in the absence of FLT3-ITD mutations. The presence of internal tandem duplications (ITD) in FLT3 is a dominant adverse prognostic factor in NK-AML, irrespective of other features. The novel method of HRM promises to revolutionise the assessment of clinical samples for both recurrent and novel mutations as it is an in-tube method that allows rapid analysis and reporting of results.

Aims
We developed assays to detect NPM1, FLT-3 ITD and FLT-3 point mutations at D835 using HRM analysis in patients with NK-AML.

Methods
DNA was extracted from archival slides from 44 newly diagnosed NK-AML patients from 1999-2007. The median age was 62 years (18-89). 31/44 (70%) were de-novo NK-AML's, 13 (30%) secondary AML's. The DNA was analysed by HRM for NPM1 and FLT-3 mutations (ITD and D835). Independent confirmatory sequencing was performed on all samples.

Results
Sixteen patients had an abnormal melting profile; 8 NPM1 alone, 4 both NPM1 and FLT3-ITD, 3 FLT3-ITD alone and 1 FLT3-D835 alone. Sequencing confirmed all HRM detected mutations and did not reveal any further mutations. The frequency of NPM mutations was lower than expected due to a moderate proportion of secondary cases. All NPM1 mutations involved a 4 base insertion and FLT3 ITD ranged from 26-102 bases. Median DFS and OS were 7 & 15 months for NPM negative/ FLT3-ITD negative patients, 13 & 14 months for NPM positive FLT-ITD negative, and 7 & 12 months for FLT3-ITD positive cases.

Conclusions
In this study, HRM was shown to be an accurate, rapid, cost-effective and efficient method of screening NK-AML samples for NPM1 and FLT-3 mutations.

Monday 15 October **0830-1000**
HSANZ Free Communications 3: Leukaemia **Meeting Room 8**

O18 0945

Analysis of the Repertoire of Immunoglobulin Heavy Chain Gene (IgH) Rearrangements in Acute Lymphoblastic Leukaemia

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2 Children's Cancer Institute Australia (CCIA), UNSW, and Centre for Children's Cancer and Blood Disorders, Sydney Children's Hospital, Sydney

Background
Determination of the detailed repertoire of immunoglobulin heavy chain gene rearrangements (IgH repertoire analysis, IRA) has recently been developed by the Flinders group as a strategy to identify molecular markers for measurement of MRD in acute lymphoblastic leukaemia (ALL).

Aim
To compare IRA to the conventional strategy developed by the European Study Group (ESG) into MRD in ALL

Methods
50 patients with B-ALL were analysed at diagnosis at CCIA using the ESG protocol, which screens a variety of immunoglobulin and T cell receptor genes for rearrangements. Two usable rearrangements were found in 39 patients, 1 in 9 patients and none in 2 patients. IgH rearrangements comprised 49% of the markers used.

Samples from these patients were analysed at Flinders by IRA. Multiple parallel Q-PCRs in microplates were used to identify the individual J and V or D gene segments participating in each IgH rearrangement. Two IgH rearrangements marking the major leukaemic clone were identified in 36 patients, 1 such rearrangement in 12 patients and none in 2 patients. IgH rearrangements marking minor leukaemic clones were observed in 28 patients; such clones made up 0.01 - 32% of the leukaemic population and were rarely detected by the ESG protocol.

Conclusions
1. IRA and the ESG protocol identify a similar total number of markers for the major leukaemic clone but IRA identifies more IgH markers. Since use of IgH rearrangements enables MRD to be quantified down to approximately 10^{-6}, identification of markers by IRA will lead to more sensitive monitoring of MRD in many patients.

2. The superior ability of IRA to detect minor clones at diagnosis, together with the potential of sensitive quantification to demonstrate early chemoresistance of such clones, may enable prospective identification of patients likely to relapse owing to the presence at diagnosis of a chemoresistant minor clone.

Monday 15 October **0830-1000**
HSANZ Free Communications 4: Myeloma I **Meeting Room 9**

O19 0830

Severe Autonomic Neuropathy Developing in Patients with Multiple Myeloma Treated with Bortezomib

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Background
Bortezomib is a proteasome inhibitor is effective in the treatment of mulitple myeloma. Postural hypotension is a frequent side effect and the underlying mechanism has not been well described. Loss of function mutations effecting the ubiquitin-proteasome system have been implicated in the pathogenesis of neurodegenerative disorders such as Parkinson's disease which is frequently complicated by autonomic dysfunction. We aimed to investigate whether bortezomib causes a significant autonomic neuropathy.

Methods and Results
Patients who commenced bortezomib for multiple myeloma during 2007 were included. Autonomic testing involved progressive head up tilt (HUT), invasive haemodynamic monitoring and determination of whole body catecholamine kinetics (noradrenaline (NA) spillover). Autonomic testing was performed after the third cycle of treatment and compared with our well established control data. Patients on bortezomib demonstrated marked reductions in noradrenaline spillover at rest and a failure to increase during orthostatic stress compared to healthy controls. The magnitude of this change is greater than that observed with centrally acting anti-hypertensives that inhibit sympathetic outflow.

	NA spillover supine (ng/min±SEM)	NA spillover 40° HUT (ng/min±SEM)	*p* value
Healthy Controls	448±45	678±76	0.008
Patients after cycle 3 Bortezomib (n=4)	204±46	255±26	0.004

We believe that the sympathetic inhibition identified in myeloma patients is unlikely to be secondary to preexisting impairment. This is supported by data for one patient who underwent baseline autonomic testing; NA spillover supine and during HUT was 365mg/min and 652ng/mi. This fell by >50% after bortezomib treatment, 204ng/min and 293ng/min. The patient suffered disabling postural hypotension.

Conclusion
Preliminary data suggest that bortezomib may result in severe autonomic dysfunction characterised by marked reductions in noradrenaline spillover and clinically significant postural hypotension. This effect appears to be reversible after cessation of the drug and is unlikely to reflect preexisting sympathetic inhibition. Targeted treatments using vasopressor agents may be useful in ameliorating postural hypotension due to bortezomib.

Monday 15 October **0830-1000**
HSANZ Free Communications 4: Myeloma I **Meeting Room 9**

O20 0845

Safety and Efficacy Results From an International Study for Expanded Access to Bortezomib: an Analysis of 109 Patients from Australia

Hang Quach[1], H Miles Prince[1], Noemi Horvath[2], Paul Cannell[3], Belinda E Butcher[4] on behalf of the Australian EAP Study Group
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Aim
The international bortezomib study in relapsed/refractory multiple myeloma (MM) was recently presented [Mikhael et al., IMW 2007]. We analysed the safety and efficacy in the Australian cohort.

Method
Eligible patients had MM with ≥2 prior lines of treatment. Patients received 1.3mg/m^2 bortezomib, IV bolus bi-weekly q3 weeks for up to 8 cycles (C). Dexamethasone, 20mg/d PO on the day of, and day after bortezomib, was added after C2 for progressive disease (PD) or after C4 for stable disease (SD). Efficacy was assessed using modified SWOG criteria; responses were defined as percent reductions in M-protein levels: Complete response (CR) - 100%; very good partial response (VGPR) 75-99%, partial response (PR) 50-74%, minimal response (MR) 25-49%, and SD, <25%. Efficacy was assessed using the intention-to-treat population. The Chi-square test compared the number of previous treatments between the Australian and international cohort.

Result
109 patients from 16 centres (55% males, median age 61.9 years) had a median of 5.2 (0-13) treatment cycles. 82% had ≥3 prior therapies. Grade 3-4 treatment related adverse events (TRAE; NCI-CTC v3) were reported in 57 pts (52%). Anaemia (19%), neutropenia (16%) and thrombocytopenia (30%) were common haematological TRAE. Herpes zoster infection occurred in 20.2%. Grade 3-4 peripheral neuropathy occurred in 3.7%. 49 patients (45%) discontinued due to AEs.

Responses were evaluable in 106 patients: 22% achieved CR/VGPR and 20% PR. Median times to first response and best response were 42 days and 69 days, respectively. Compared to the international data, the Australian cohort demonstrated an inferior overall response rate (42% vs. 54%, p=0.006), possibly due to heavier pretreatment (82% had ≥3 prior therapies vs. 68%, p<0.001).

Conclusion
Bortezomib demonstrates high efficacy even in heavily pre-treated MM patients; this analysis confirms that bortezomib is more efficacious in less heavily pre-treated patients. TRAE were common, but were manageable.

Monday 15 October **0830-1000**
HSANZ Free Communications 4: Myeloma I **Meeting Room 9**

O21 0900

Bortezomib-Associated Thrombocytopenia is Reduced by Concurrent Use of Dexamethasone

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Aim
Bortezomib-related thrombocytopenia is a well recognized adverse event. We examined the effect on this phenomenon of the concurrent administration of dexamethasone (40mg per week or equivalent dose of corticosteroids).

Method
A retrospective analysis was performed on 34 patients with relapsed or refractory multiple myeloma, treated with bortezomib (1.3mg/m2 IV bi-weekly q3 weeks) ± dexamethasone (20mg/d on day of and day after bortezomib) at Peter MacCallum Cancer Centre between Februrary 2005 to February 2007. Data were abstracted regarding patients' baseline characteristics, platelet count at the beginning, at nadir, and the percentage of platelet reduction as well as the number of platelet transfusions with each treatment cycle. Comparisons of these parameters between the two groups were performed using the two-sample t-test.

Results
34 patients received a median of 4 cycles (range 1-10) of bortezomib ± dexamethasone; 16 and 18 patients had a total of 75 cycles with dexamethasone and 65 cycles without dexamethasone, respectively. Both groups had similar baseline characteristics including mean platelet count prior to cycle 1 (196 x 10^9/l vs.160x10^9/l; p=0.27). Patients who received dexamethasone had a higher mean platelet count at the start of each new cycle (217x10^9/l vs. 171x10^9/l; p=0.002), a higher nadir platelet count (149x10^9/l vs. 88x10^9/l; p<0.001) and a lower mean platelet reduction (33% vs. 51%; p<0.001) per treatment cycle. Patients who did not receive dexamethasone had a higher average number of platelet transfusions per treatment cycle (0.86 vs.0.05, p=0.004).

Conclusion
Bortezomib treatment induces a mean platelet reduction of approximately 50%, which is comparable to previous published reports. Dexamethasone reduces the incidence and depth of bortezomib-associated thrombocytopenia with subsequent reduction in platelet transfusion requirements. Further studies are required to confirm this observation and investigate the underlying mechanisms. We recommend the concurrent use of dexamethasone for patients with low baseline platelets being treated with bortezomib.

Monday 15 October **0830-1000**
HSANZ Free Communications 4: Myeloma I **Meeting Room 9**

O22 0915

VEGF-R Inhibition with Pazopanib (GW786034) is Ineffective in Pretreated Myeloma

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Aim
Pazopanib (GW786034) is a potent, small molecule inhibitor of VEGFR-1, 2, and 3, PDGFR-alpha and beta, and c-kit which has demonstrated clinical activity in renal cell carcinoma, ovarian cancer, sarcoma and other solid tumors. We aim to evaluate the safety and efficacy of Pazopanib in relapsed and refractory multiple myeloma (MM).

Methods
This was an open-label, single arm phase II trial in patients with relapsed/refractory MM. The primary objective was response rate, and secondary objectives were time to MM progression, pharmacokinetics (PK) and toxicity. A 2-stage Green-Dahlberg design was utilized with provision for early termination if < 3 responses (Blade criteria) were reported in Stage 1 (n=20). Patients received 800mg pazopanib p.o. daily.

Results
21 pts with MM were treated: median age 59 yrs (range 29-74); SWOG stage 1/2 (71%). All pts were heavily pre-treated, including 86% with ≥4 prior chemotherapy regimens and 71% with prior stem cell transplantation. Treatment duration ranged from 14 to 279 days (mean= 69 days). No clinical responses were observed in 19 evaluable pts. 18 patients (86%) were discontinued from study due to disease progression; two (10%) were discontinued due to toxicity (Gr 2 fatigue; Gr 3 nausea/vomiting) and one patient was a protocol violation completing one dose of pazopanib. The most common drug-related toxicities (n=21 pts) were nausea (38%), fatigue (29%), hypertension (24%), epistaxis (19%), headache (19%), and hair color changes (19%). No venous thromboembolism occurred . No adrenal dysfunction was apparent. PK data paralleled the data observed in other pazopanib trials where responses in renal cell carcinoma, ovarian cancer, sarcoma and other solid tumours have been observed.

Conclusion
Despite using a dose that has demonstrated activity in various solid tumours, and achieving similar PK profiles, with demonstrable clinical 'biomarker' activity (hair colour change, hypertension), pazopanib did not demonstrate clinical activity in treatment of MM.

Monday 15 October **0830-1000**
HSANZ Free Communications 4: Myeloma I **Meeting Room 9**

O23 0930

The Epidemiology of Clonal Lymphocytosis of Undetermined Significance (CLUS) in Australia

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Aim
To identify the frequency and nature of small clonal populations in Australia.

Methods
A retrospective analysis of blood count, flow cytometry and referral data was conducted at a large, non-hospital based pathology laboratory in Sydney, Australia servicing both metropolitan and regional areas of New South Wales using the criteria of Marti et al, BJHaem, 2005.

Results
There were 414 patients identified from 2000 to 2005 with an incidental finding of a B-cell clone with total B-lymphocytes $<5.0x10^9$/L and no adenopathy, organomegaly, cytopenia or clinical features of lymphoproliferative disorder. There were 212 males (51%) and 202 females (49%) with a mean age of 69.7 years (range of 14-97 years). Patients were divided into two groups: 322 (77.7%) with a typical CLL phenotype and 92 (22.2%) with a 'non-CLL' phenotype.

Of patients with a CLL phenotype, 168 (52.2%) were male and 154 (47.8%) female. No patient had a CLL clone younger than 40 years, and for each subsequent age decade there were 4% (40-49 yrs), 13% (50-59yrs), 23% (60-69yrs), 40% (70-79yrs), 18% (80-89yrs) and 2% (>90yrs) of the patients with a CLL clone. The mean clonal level (CD19/CD5+) was $2.36x10^9$/L (range 0.01-$5.21x10^9$/L) and the absolute lymphocyte count (ALC) was 0.4-$10.5x10^9$/L. Only 156 (48%) of these patients do not fulfil NCI-1996 criteria for the diagnosis of CLL (ALC $<5.0x10^9$/L), 71 (22%) had an ALC within the laboratory reference range (1.0-4.0 $x10^9$/L).

Analysis of the 92 patients with a 'non-CLL' phenotype showed 44 (47.8%) males and 48 (52.2%) females. There were 65 (70.6%) with a phenotype of 'lymphoma' (otherwise unspecified) and the remainder had more specific phenotypes. The mean clonal absolute count in this group was $1.27x10^9$/L.

Conclusions
CLUS / MBL is not uncommon in the immunophenotyping laboratory as an incidental finding. The majority have a phenotype typical of CLL while there are a range of non-CLL phenotypes identified, the latter in particular of uncertain significance in patients with no other clinical findings.

Monday 15 October **0830-1000**
HSANZ Free Communications 4: Myeloma I **Meeting Room 9**

O24 0945

Prevalence of MGUS/Myeloma in Patients Presenting with Acute Osteoporotic Vertebral Fractures over 18 Months

Terry Diamond and Terry Golombick
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Aim
To determine the prevalence of MGUS/myeloma in patients presenting with acute osteoporotic vertebral fractures over 18 months.

Design
Retrospective, cross sectional, observational study.

Methods
We studied 134 consecutive patients with acute vertebral fractures (defined by MRI criteria) presenting over an 18 month period to our osteoporotic clinics. All patients had densitometric evidence of osteoporosis. Patients were grouped according to their underlying cause of osteoporosis (postmenopausal, glucocorticoid induced, multiple myeloma, monoclonal gammopathy of undetermined significance and other). Their data comprised of clinical demographics (age, sex, smoking history, alcohol intake), serum biochemistry (calcium, 25OHD3, PTH), bone densitometry (DXA and QCT), markers of bone resorption (u-DPYD) and spinal radiology. Patient data were expressed as mean $\pm$ 1 SEM and compared using ANOVA.

Results
There were 106 women and 28 men, with mean age of 78.4 years. 57% (n=77) were defined as having postmenopausal osteoporosis, 14.9% (n=20) as MGUS and 4.5% (n=6) as multiple myeloma. On average, mean lumbar spine BMD T-score value was –3.4 with each patient having 4.2 prevalent vertebral fractures. No difference in severity of osteoporosis, total number of vertebral fractures or clinical variables was noted in the 5 groups. Patients with plasma cell disorders had a mean paraprotein of 6.4g/L and a mean Hb of 132. Multiple regression analysis demonstrated that vitamin D was the most important predictor of vertebral fractures ($P<0.0001$) and weight of LS BMD ($P<0.0001$), after controlling for other variables.

Conclusions
Previous findings suggest that 5% of newly diagnosed patients presenting with densitometric evidence of osteoporosis have MGUS or myeloma. To our knowledge, this study is the first to evaluate the prevalence of these dyscrasias in patients with acute vertebral fractures. Our data demonstrate that MGUS and myeloma may account for up to 20% of an acute vertebral fracture cohort.

Monday 15 October **0830-1000**
ANZSBT Presidential Symposium **Central Room A**

O25 0830

Challenges of Rhesus D Prophylaxis

Bernie Ingleby, Chris Arnold, Andrew Barr, Catherine Cole
PathWest, Women's and Children's Health Service; King Edward Memorial Hospital (KEMH) and Princess Margaret Hospital, Perth, Western Australia

Revised ANZSBT guidelines were recently released in response to two adverse events in Australia where immune anti-D was confused as Rh(D) Immunoglobulin (RhIG). They attempt to reduce this risk by advising anti-D be reported as "suggestive of an immune response" requiring titre and referral to specialist when:

RhIG history unknown or given >6 weeks ago.
RhIG <6 weeks ago with reaction 2-4+ (grading 0-4).
However, patients with immune anti-D are not given RhIG.
KEMH is Western Australia's only tertiary maternity hospital with 5,700 births/year. The Transfusion Medicine Unit (TMU) reviewed 95 samples with detectable RhIG and found:

When RhIG <6 weeks ago, 51% (37 of 73) gave 2-4+ reaction.
17% (15 of 88) were detected 6-12 weeks after injection.
7% (7 of 95) had unknown history but timing suggested antenatal RhIG given elsewhere.
Overall, 67% would be incorrectly reported as immune anti-D increasing the risk that RhIG would be discontinued with unnecessary referral and monitoring.
KEMH guidelines offer a better alternative where:

Local validation by AutoVue Innova® provides a benchmark for expected post-RhIG results:
Report as likely RhIG when given <6 weeks ago with reaction 1-3+, or when RhIG given 6-12 weeks ago giving reaction 1-2+.
Report as immune when RhIG given <6 weeks ago with reaction 4+, when RhIG given 6-12 weeks ago giving reaction 3-4+, or when RhIG given >12 weeks ago. Titre and refer to specialist.
RhIG history unknown, report as immune if titre ≥1:8 (refer to specialist), or ?RhIG/Immune if titre <1:8 (monitor).
Patients attend KEMH for antenatal RhIG.
TMU takes active role by issuing RhIG, ensuring collection of pre-RhIG samples, providing helpful report comments, liasing closely with hospital staff, checking maternal record for all cord samples, and following up if RhIG not requested.

Monday 15 October **0830-1000**
ANZSBT Presidential Symposium **Central Room A**

O26 0845

Bloodless Medicine, Surgery and Stem Cell Transplantation at the Center for Bloodless Medicine and Surgery, Pennsylvania Hospital, Philadelphia, USA

Ruth Power
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Aim
To develop knowledge relating to appropriate blood transfusion in the context of bloodless medicine.

Method
As the recipient of the ANZSBT / CSL Bioplasma Overseas Travel Grant 2006, I utilised the grant to fund an educational visit to the Center for Bloodless Medicine and Surgery (CBMS) at Pennsylvania Hospital in April 2007.

Results
The CBMS offers medical and surgical alternatives to patients, such as Jehovah's Witnesses, unwilling to accept transfusion of blood or blood products. Bloodless care by the CBMS incorporates 6 basic building blocks of blood management which are important across the entire patient population but critical in the anaemic patient. They are applied to medical, surgical and transplant patient throughout the entirety of the patient's care. For CBMS patients, anaemia is managed using prophylactic erythropoietin, IV iron therapy and volume support, and thrombocytopenia is managed using haemostatic agents, vitamin K and interleukin-11.

Of the 128 patients with profound anaemia (Hb < 70g/L) treated without blood by the CBMS between January 2002 and August 2005, the survival rate was 95.3% (N=122) with 100% survival for Hb 51-70g/L (N=78) and 88% survival for the 50 patients with Hb 25-50g/L(N=44).

The Bloodless Stem Cell Transplant Program at the CBMS offers bloodless stem cell transplantation for patients diagnosed with lymphoma or multiple myeloma, and bloodless treatment for acute leukaemia patients. To date, the CBMS had performed 55 bloodless autologous peripheral blood stem cell transplants (PBSCTs) with 2 mortalities (96.4% survival) and one serious bleeding complication. Two patients with AML have been treated by the CBMS without transfusion. One survived standard induction chemotherapy and one cycle of remission consolidation chemotherapy prior to relapse. The second survived reduced induction chemotherapy and 4 cycles of remission consolidation chemotherapy but relapsed after 12 months. Future goals for the CBMS include further development of strategies for bloodless treatment of acute leukaemia and the implementation of a bloodless allogeneic PBSCT Program.

Conclusion
Predetermined transfusion triggers may not be the best indicator for appropriate transfusion. Individual patient needs and the patient's clinical response are better markers. Profound anaemia and autologous PBSCT with high dose chemotherapy can be managed safely without transfusion.

Monday 15 October **0830-1000**
ANZSBT Presidential Symposium **Central Room A**

O27 0900

Microparticles in Stored Platelet Concentrates and their Immunomodulatory Potential

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Aim
Microparticles are cell-derived fragments that accumulate in stored blood transfusion products, including platelet concentrates (PCs). The pro-coagulant properties of platelet-derived microparticles in various disease states have been recognised for some time, but little is known about their potential to illicit an immune response. Furthermore little is known about the bioactive potential of microparticles generated during storage of PCs. Our aims were to determine the contribution of microparticles to the platelet storage lesion, to assess whether microparticles stimulate leucocytes and to identify bioactive proteins present on microparticles.

Method
Leucodepleted PCs were prepared and stored according to standard blood bank procedures. Supernatant samples were collected throughout storage. Half of each supernatant sample was ultracentrifuged to fractionate the microparticles and supernatant. A variety of techniques including ELISA were used to examine bioactive proteins present in supernatant and fractionated samples. Microparticles were characterised by flow cytometry for platelet markers including CD42b and CD41. An *in vitro* 'responder' model of transfusion involving the incubation of allogeneic leucocytes with stored supernatant (± microparticles) was used to determine leucocyte activation and subsequent cytokine production.

Results
Storage-generated microparticles contain platelet-derived bioactive proteins including CXCL7, PDGF, EGF, sCD40L, CCL5 and BDNF, but the concentrations of these factors were lower in microparticles than the corresponding microparticle-depleted supernatant. Storage time showed a positive correlation with increased leucocyte response. PC supernatant induced greater activation of 'responder' leucocytes than the corresponding microparticle fraction.

Conclusion
Storage generated microparticles contain bioactive proteins indicative of platelets. Microparticles showed lower potential for stimulating allogeneic leucocytes than microparticle-depleted supernatant, suggesting that truly soluble factors are responsible for this effect. The potential of microparticles present in stored PCs to influence other clinically-relevant biological processes in the transfusion recipient requires further investigation.

Monday 15 October **0830-1000**
ANZSBT Presidential Symposium **Central Room A**

O28 0915

Buffy Coat Pooled Platelets Stored in SSP+ for 7 Days Show Superior in vitro Function to Day 5 Platelets Stored in T-Sol

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Aim
Third generation platelet additive solutions such as SSP+ offer improved conditions for platelet support during storage. As bacterial detection and reduction techniques improve, focus is moving toward extending the shelf life of both buffy coat pooled and apheresis platelets. Previous studies have shown improved in vitro function of apheresis platelets stored in SSP+ (MacoPharma). The aim of this study was to investigate whether buffy coat pooled platelets stored for seven days in SSP+ retained superior function compared to paired buffy coat platelets stored in T-Sol (Baxter).

Methods
Buffy coat pooled platelets (n= 12) were prepared, split and resuspended at equivalent concentrations in either SSP+ or T-Sol. On days 1, 6 and 8 following collection, platelets were tested for pH, platelet count, hypotonic shock response (HSR), extent of shape change in response to ADP (ESC), and expression of activation markers CD62P and CD63. In all cases platelets were resuspended in AB plasma prior to assessment of functional response. Results were analysed using paired T-tests.

Results
Volume, platelet count, pH and white cell contamination were not significantly different on day 1. By day 6 and 8 platelet count was significantly higher when stored in SSP+ ($p=0.02$, $p=0.008$ respectively). Function as determined by HSR and ESC was significantly higher in platelets stored in SSP+. At days 6 and 8 HSR was 45 and 40% for SSP+ platelets compared to 27% and 23% for T-Sol platelets ($p<0.005$ in both cases). Functional activity correlated well with activation status. Platelets stored in T-Sol were significantly more activated than platelets stored in SSP+ at both days 6 (37% vs. 10%; $p<0.001$) and 8 ($p=13.8\%$ vs. 38.6%; $p<0.001$).

Conclusions
Both platelet survival and In vitro assessment of function suggests that platelets stored for 7 days in SSP+ are functionally superior to platelets stored in T-Sol for 5 days.

Monday 15 October **0830-1000**
ANZSBT Presidential Symposium **Central Room A**

O29 0930

Duration of Packed Cell Storage is Associated with Hypercoagulable Thromboelastogram Changes in an *in vitro* Model of Transfusion

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Red cell apoptosis is characterized by loss of lipid asymmetry and cleavage of trans-membrane and cytoskeletal proteins by proteases such as caspase 3. We have previously demonstrated that increased red cell susceptibility to apoptotic change is associated with duration of packed cell storage as evidenced by increased Annexin V binding of exposed phosphatidylserine (PS). We found a 12.6 fold increase in Annexin V binding to red cells after 42 days storage. We postulated that transfusion of aged packed cells with increased apoptosis and exposure of PS, may induce a hypercoagulable state.

We utilised the thromboelastogram to assess coagulation changes in an *in vitro* model of transfusion. Fresh whole blood samples were collected from volunteers and samples spiked with either N/Saline or packed red cells which had been stored for variable time periods (Day 11-122). Samples were spiked to model 10%, 25% and 50% transfusions. TEG results were compared to an unspiked baseline sample to allow for variation between donors. Haematocrit levels were measured on Coulter LH750.

For packed cells stored up to 34 days prior to testing, TEG results were not significantly different when compared with NSaline spiked samples. For packed cell samples stored longer, there was a shortening of R time and K time and an increase in MA (max amplitude) consistent with a hypercoagulable state. These changes were most apparent in the model of 50% transfusion but were also seen in 10% and 25% models. Variations in haematocrit alone did not account for the hypercoagulable changes seen.

In our *in vitro* model of transfusion, duration of packed cell storage was associated with demonstration of hypercoagulable changes utilising the TEG. Whether these changes may be prevented by leucodepletion is the subject of ongoing work.

Monday 15 October **0830-1000**
ANZSBT Presidential Symposium **Central Room A**

O30 0945

An Assessment of the Risk of Transfusion Acquired CMV (TA-CMV) in Australia From Seronegative Blood Components

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Australian Red Cross Blood Service

Aim
TA-CMV is minimized by (1) donor screening for specific antibody (supporting its issue as 'CMV seronegative') and/or (2) leucodepletion of blood components (since CMV is a leucocyte-associated virus). Few data exist regarding the risk of TA-CMV in Australia. We analysed the TA-CMV risk associated solely with non-leucodepleted blood components issued as CMV seronegative.

Methods
The rate that CMV seronegative blood donors (n=5,140) in South Australia 'seroconverted' to become CMV antibody positive was analysed. The CMV antibody window period (WP) was derived using 13 seroconversion panels sourced from pregnant women. A published model (incidence rate/WP) was used to estimate the probability that a donor with primary CMV infection donated during the WP. The average probability of CMV DNA presence during seroconversion was calculated from two studies of seroconverting blood donors as 0.011. The risk of TA-CMV was assessed as the product of these two probabilities i.e. P (WP) x P (viremia).

Results
Twenty eight (28) of the 5,140 (0.54%) previously CMV antibody negative donors seroconverted. The median time to first detection of CMV antibody was day 46, designated as the antibody WP. The probability of a WP donation calculated as 68.65×10^{-5} or 1 in1,457. The risk of releasing a CMV seronegative blood component containing CMV DNA was estimated as 7.895×10^{-5} or 1 in 126,661.

Conclusion
This analysis suggests that the risk of TA-CMV associated with seronegative non-leucodepleted blood components in Australia is low. Based on the risk estimate and considering that the ARCBS issued almost 138,000 CMV seronegative components in 2006/07, it might be expected that Australia wide approximately one potentially infectious WP unit could be issued per annum. However, the likelihood of infection in the recipient will depend on various factors including component viral load and leucodepletion status, and recipient immune status.

Monday 15 October **0830-1000**
ASTH Free Communications 1: Laboratory Science/Techniques
Central Room C

O31 0830

Influence of Lepirudin and Warfarin on APTT, Ecarin Clotting Time and INR: Implications for Laboratory Monitoring of Patients with Heparin Induced Thrombocytopenia

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Lepirudin is indicated for the acute management of heparin induced thrombocytopenia (HIT). Two clotting assays, the activated partial thromboplastin time (APTT) and the ecarin clotting time (ECT) can be used to monitor lepirudin. The APTT is a widely available and familiar measurement to most laboratories and clinicians, however underestimates higher lepirudin plasma levels. The ECT is a more linear measure however is not so commonly utilized. To determine an optimal laboratory strategy to monitor lepirudin before and during warfarin therapy we have examined the influence of these anticoagulants on the APTT, ECT and INR in vitro and in patients treated for HIT. Pooled normal plasma and pooled plasma from patients on warfarin were spiked with lepirudin and clotting assays were performed on the Diagnostica Stago STAR analyser. In vitro, APTT and ECT were more prolonged in the presence of warfarin and lepirudin compared to lepirudin alone. This effect was less marked for ECT compared to APTT (18% and 37% increase respectively) in pooled INR 2.4 plasma at 0.5ug/ml lepirudin. Secondly, INRs from patients on warfarin were further prolonged in vitro by the addition of lepirudin (29%-58% increase for INRs 1.9 to 3.0 at 1.0ug/ml lepirudin). Corresponding patterns were observed in patients on lepirudin and warfarin therapy. In summary warfarin and lepirudin have synergistic effects on APTT, ECT and INR which can lead to uncertainty of anticoagulant status during lepirudin-warfarin cross-over. While the ECT is a more accurate measure of lepirudin it is also affected by the addition of warfarin although to a lesser extent than the APTT. In addition, estimating a therapeutic warfarin dose while on lepirudin needs to take into account up to 30-50% inflation of INR due to lepirudin. These complex interrelationships need to be considered in designing clinical algorithms for monitoring lepirudin to warfarin anticoagulation in HIT.

Monday 15 October **0830-1000**
ASTH Free Communications 1: Laboratory Science/Techniques
Central Room C

O32 0845

A Rapid Automated Technique to Detect Platelet Activation in Plasma

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[1]Royal Perth Hospital, Perth, Western Australia' [2] Princess Margaret Hospital, Toronto, Canada [3] McMaster University, Hamilton, Canada

Aim
To compare an automated technique for the measurement of plasma procoagulant phosphophlipid (PPL)(XACT test, Haematex Research, NSW, Australia) with more established laboratory tests of platelet activation and aggregation. The XACT test measures the clotting time for recalcified plasma after the addition of FXa.

Methods
The STA-R coagulation analyser (Diagnostica Stago, Asnsières, France) was used to perform the XACT test. Plasma soluble CD40 Ligand (sCD40L) was performed by ELISA (Bender MedSystems, CA, USA). Optical platelet aggregation with ADP at 10 μmol/L was performed on the Chronolog 680 analyser (Chronolog, PA, USA) and whole blood aggregation with ADP on the PFA 100® analyser (Dade Behring, Germany). Urinary 11-dehydro thromboxane B_2 (11 dehydro TxB_2) (Cayman Chemical Company, MI, USA) was performed by ELISA. A total of 450 samples were tested with most patients having paired pre and post percutaneous coronary intervention (PCI) collections. All patients received aspirin and clopidogrel and two thirds received GPIIb/IIIa inhibitors.

Results
The results for the paired pre and post PCI are shown in table 1 (Mean +/- SD). There was a significant decrease in PPL post PCI consistent with platelet inhibitors administered peri-procedure.

Table 1

	PPL (μg/mL)	sCD40L (ng/mL)	11-dehydro TXB_2 (pg/mmol creatinine)
Pre	1.48(1.21)	1.85(2.17)	72.89(46.68)
Post	1.07(1.19)	1.87(2.32)	82.75(64.42)
Significance	<.0001	0.837	0.0182
Paired number	202	200	192

Overall there was correlation between the XACT test and sCD40L (r = 0.1395, P =0.0032). There was no significant correlation with platelet aggregation, PFA 100® or urinary 11- dehydro TxB_2 (Table 2).

Table 2

	sCD40L	11- dehydro TxB2	ADP 10 μmol/L	PFA 100®
PPL	r = 0.1395 (P =0.0032)	r = -0.0335 (P = 0.4879)	r =-0.0270 (P = 0.7355)	r = -0.0842 (P = 0.2931)

Conclusion
The XACT test is a rapid, simple and cost-effective way to measure PPL. It was significantly lower after PCI, most likely due to the administration of anti platelet drugs. In a large group of patients it correlates with sCD40L, a more established marker of platelet activation but not with other laboratory measures of platelet function.

Monday 15 October **0830-1000**
ASTH Free Communications 1: Laboratory Science/Techniques
Central Room C

O33 0900

Comparison of the VASP Flow Cytometric Assay and *Verify Now* P2Y12 Inhibition Assay for the Assessment of Clopidogrel Responsiveness

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Introduction

Inter-patient variability in response to Clopidogrel has been reported and necessitated the development of assays for detecting individual responsiveness. The *Biocytex* VASP flow cytometric assay measures the phosphorylation of intracellular VASP (Vasodilator Stimulated Phosphoprotein) in response to platelet ADP stimulation. The *Accumetrix Verify Now* assay is a functional assay that measures platelet P2Y12 inhibition following ADP stimulation.

Aim

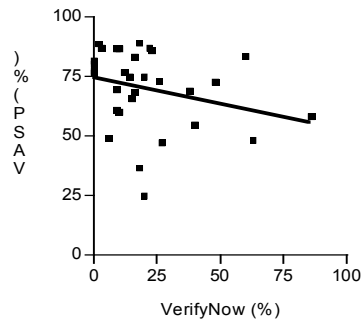
The aim of this study was to correlate the VASP and Verify Now assays and to investigate the individual responsiveness of patients undergoing coronary artery stenting to Clopidogrel.

Method

Whole blood samples were obtained from 31 patients (17M/14F) undergoing coronary artery stenting after receiving a loading dose of 600mg Clopidogrel more than 3 hours prior to blood collection. Simultaneous samples were tested for VASP and Verifynow and VASP was also measured in 5 normal controls.

Results

There was a significant inter-patient variability in the responsiveness to Clopidogrel as detected by both VASP and Verify Now assays, ranging from 24.7% to 88.8% and 0.0% and 86.8% respectively. Mean VASP phosphorylation was significantly decreased in clopidogrel-treated patients (70.1%) compared to controls not receiving clopidogrel (87.7%, p=0.025). Despite the inter-patient variability detected by both assays, there was little correlation between VASP phosphorylation and Verify Now P2Y12 inhibition (p=0.13) as shown.



Conclusions

Individual patient responsiveness to Clopidogrel was detected in patients undergoing coronary stenting using both VASP and Verify Now assays. However, there was no significant correlation between the results of these two assays suggesting that factors apart from P2Y12 inhibition alone may influence these tests.

Monday 15 October **0830-1000**
ASTH Free Communications 1: Laboratory Science/Techniques
Central Room C

O34 0915

Poor Correlation Between Activated Whole Blood Clotting Time (ACT) and AntiXa Levels in Patients Undergoing Coronary Artery Bypass Graft Surgery (CABGS)

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Department of Anaesthetics, Royal Melbourne Hospital, Parkville VIC

Background
Haemostatic regulation and monitoring plays an integral role during "on-pump" CABGS. A critical balance exists between adequate anticoagulation and excessive haemorrhage. The ACT is commonly employed as a point-of-care test of coagulation to guide heparin administration and reversal with protamine. There is, however, debate in the literature pertaining to its reliability.

Aim
To evaluate the reliability and utility of the ACT method for monitoring coagulation during "on-pump" CABGS employed in our institution, and to assess the correlation between these measurements and the doses of heparin and protamine given.

Methods
Blood samples from 15 consecutive patients undergoing "on-pump" CABGS were assayed pre-bypass, regularly during bypass and post heparin reversal by protamine, using 3 point-of-care ACT instruments (Medtronic®, Acatalyke® and Istat®) and a chromogenic AntiXa assay (STA®). Heparin and protamine doses and postoperative blood product usage were recorded.

Results
The mean total heparin dose was 485.6units/kg±160units/kg. Considering all ACT values there was significant correlation between the Medtronic® and Acatalyke® (r=0.88 p=<0.0001) and Istat® (r=0.91 p<0.0001). The correlation, however, between respective instruments was significantly diminished at higher ACTs (>450) required on bypass (>400sec – Medtronic® vs Acatalyke® r=0.63 P<0.0001; Medtronic® vs Istat® r=0.73 P<0.0001 and >600sec – Medtronic® vs Acatalyke® r=0.27 P=0.12; Medtronic® vs Istat® r=0.447 P=0.006). The correlation between antiXa levels and ACT was moderate when all values were considered (Medtronic® r=0.77 P<0.0001; Acatalyke® r=0.79 P<0.0001 and Istat® r=0.80 P<0.0001). However at therapeutic heparin levels, the correlation was abolished (post heparin - Medtronic® r=0.34 p=0.006; Acatalyke® r=0.41 p=0.001 and Istat® r=0.49 P<0.0001). The mean protamine dose expressed in terms of heparin dose (mg/units heparin) was 0.92mg/1000Units ± 0.42mg. The dose of protamine, however, failed to strongly correlate with either the antiXa (r=0.57 p=0.027) or ACT (r=-0.24 p=0.381). Post-operatively 53% of patients required transfusions, but there was no significant correlation with any of the measured parameters.

Conclusion
There is poor concordance between ACT measurements performed on different instruments at heparin doses necessitated by bypass. The correlation between ACTs and heparin concentrations derived from antiXa levels is weak. Further, neither the dose of heparin or protamine appear to be standardised.

Monday 15 October **0830-1000**
ASTH Free Communications 1: Laboratory Science/Techniques
Central Room C

O35 0930

Utility of Platelet Aggregation Studies in Patients with a Positive Immunoassay Test for HIT Antibodies in a Tertiary Hospital Setting

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Background
Immunoassays to detect HIT antibodies have significant false positive rates, especially in patients with clinically defined low and intermediate pre-test probabilities for the syndrome.

Aim
A retrospective audit to determine the utility of platelet aggregation studies in patients with a positive immunoassay test for HIT antibodies was performed.

Methods
All patients with a positive HIT antibody via the Diamed or ELISA immunoassay test from May 2005 to May 2007 were included for analysis. Their clinical records were reviewed and a "pre-test" probability according to the 4 T's scoring system was assigned. Platelet aggregometry results were used as a functional test, serving as a surrogate marker for heparin induced platelet activation.

Results
127 patients were tested using the Diamed or ELISA assay and 22 had a positive HIT antibody detected. The group consisted of surgical (n=12) and medical patients (n=10). The pre-test probability was high in 18% (4/22); intermediate in 50% (11/22) and low in 32% (7/22) of patients. All patients had heparin ceased with 20 receiving alternative anti-coagulation (danaparoid/bivalirudin/lepirudin). There were 4 thrombotic events, 3 in the high and 1 in the intermediate probability group. All patients with a high pre-test probability had positive platelet aggregation, as did 10/11 in the intermediate group. 6/7 in the low probability group were tested for platelet aggregation; 3 were positive with 1 indeterminate result.

Conclusion
In this small cohort, platelet aggregometry does not appear to be a useful adjunct to immunoassays in the diagnosis of HIT, particularly in the intermediate probability group where a definitive test would be most helpful. Further correlation will be undertaken with the serotonin release assay.

Monday 15 October **0830-1000**
ASTH Free Communications 1: Laboratory Science/Techniques
Central Room C

O36 0945

Use of 4T's Score and Diamed PaGIA to Guide Management of HIT - A Single Institution Experience

Adam Bryant, Joyce Low, Steve Austin, Joanne Joseph
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Introduction
Heparin induced thrombocytopenia (HIT) remains an important clinical entity. Although serotonin release assay (SRA) is considered the 'gold standard' diagnostic test, it is generally necessary to institute therapy for HIT prior to the SRA result being known.

Aim
In a cohort of 199 suspected HIT cases, we have previously demonstrated that all patients with a low pre-test probability or negative Diamed PaGIA Heparin/PF4 antibody test (PaGIA) had negative SRA (NPV 100%). Although all patients with positive SRA also had a positive PaGIA, the PPV of PaGIA was only 39%. The aim of this current work was to review progress and outcome of all patients with positive PaGIA, particularly with respect to its influence on clinical management.

Results
The relevant details of 18 patients with positive PaGIA are tabulated and grouped according to 4T's PTP score. After review of records, progress and all results, a final diagnosis of HIT was assigned as probable, possible or unlikely.

4T's PTP score	Number of patients	SRA result	Alternative a/c commenced	Final HIT diagnosis
High	5	4/5 positive	4/5 danaparoid	3/5 probable 2/5 possible
Intermediate	8	3/8 positive	3/8 danaparoid	2/8 probable 2/8 possible 4/8 unlikely
Low	5	0/5 positive	1/5 danaparoid	5/5 unlikely

Conclusion
Whilst diagnosis of HIT remains difficult, it appears prudent to manage PaGIA positive patients who have an intermediate or high score with alternative anti-coagulation. No PaGIA positive patients with a low score were ultimately considered to have HIT, thus it may be reasonable to continue these patients on unfractionated heparin, particularly if switching to an alternative anticoagulant may be problematic.

Monday 15 October **0830-1000**
Nursing Free Communications 1 **Meeting Room 5**

O37 0830

Granulocyte Collections – One Unit's Experience

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Aim
An apheresis unit's recent experience in the collection of donor granulocytes has been reviewed. The objective of the review was to identify variables that may affect the quality of the collected product.

Method
Review of relevant literature and procedural records of 32 donations was performed. Variables identified included:
Granulocyte stimulating agents for donors
Type of cell separator used
Procedural problems – donor and cell separator
Pre-donation granulocyte counts
Blood volume processed
Use of red cell sedimenting agents
Collection results

Result
Therapeutic doses of granulocytes were collected from 30 of the 32 donation episodes. Decrease in collection yield may have been related to a decrease in blood volume processed in one episode, and a low donor white cell count pre granulocyte donation in the other

Conclusion
In this unit's experience, a therapeutic dose of granulocytes can be collected if the donor's white cell count is greater than 20^9/L, a red cell sedimenting agent is used, more than one blood volume is processed, and there are no issues that affect the interface of the cell separator.

Monday 15 October **0830-1000**
Nursing Free Communications 1 **Meeting Room 5**

O38 **0845**
Donor Lymphocyte Infusion (DLI) – What and Who?

Terri Larkin-Andelkovic, Ron Middleton, Maree Bransdon, Alanna Geary

Aim
Explain what is a DLI? Who receives DLI and why?

Method
Literature review of scientific and medical journals to develop and implement a facility policy for the administration of DLI

Results
Donor Lymphocyte Infusions (DLI) is an infrequent treatment for patients post allogenic Stem Cell Transplant (SCT). Donor Lymphocytes are collected using the same apheresis principles as Peripheral Blood Stem Cells (PBPC). If a DLI is required enough cells are collected for four infusions.

The majority of the papers reviewed discussed the medical and scientific aspects related to DLI's. The literature was lacking information relating to the administration, nursing management and care for patients receiving DLI's. To ascertain if there was any current information and/or policies/protocols related to the administration, nursing management and care for patients, other hospitals who might potentially administer DLI's and members from the Transplant Nurses Association were contacted. Unfortunately this avenue did not provide any information on this topic, and none of the avenues explored could provide policies/protocols.

As there is little information on the administration, nursing management and care of DLI's it was evident that a written policy was required. The DLI policy that has been developed aims to provide information, general guidelines, potential complications and safe administration of DLI's in the outpatient setting for nursing staff within Cancer Care Services at the Royal Brisbane and Women's Hospital.

Monday 15 October **0830-1000**
Nursing Free Communications 1 **Meeting Room 5**

O39 0900

Therapeutic Plasma Exchange for Goodpasture's Syndrome with Negative Anti-GBM Antibody

Kari Mudie, Glen Kennedy, Maree Bransdon, Alanna Geary
Royal Brisbane & Women's Hospital, Brisbane, Queensland

The introduction of therapeutic plasma exchange and immunosuppressants as standard treatment decreased the high mortality rate associated with Goodpasture's Syndrome. Goodpasture's Syndrome is a rare autoimmune disorder that affects the glomerular basement membrane of the kidneys and/or the alveolar basement membrane of the lungs.

The acute presentation of a 22yr old woman (Ms T) diagnosed with Goodpasture's Syndrome required immediate commencement of standard treatment. The patient presented with all the classic symptoms: acute renal failure, pulmonary haemorrhage and positive serum anti-glomerular basement membrane antibodies. Patient response to standard treatment was favourable and the patient was discharged home.

Ms T re-presented 2 weeks later, again with pulmonary haemorrhage and acute renal failure, however, on this occasion the patient had a negative serum anti-glomerular basement membrane antibody assay, not commonly seen in acute stages of Goodpasture's Syndrome. The response to standard treatment regime on this occasion was not favourable.

The case study will compare the efficacy of therapeutic plasma exchange for Ms T in both the presence and absence of anti-GBM antibody. The presentation will discuss the impact of contributory factors associated with Ms T's case. The provision of care for Ms T demonstrated the need for educational tools across specialities.

Monday 15 October **0830-1000**
Nursing Free Communications 1 **Meeting Room 5**

O40 **0915**

Therapeutic Plasma Exchange for Transplant Related Thrombotic Thrombocytopenia Purpura – RBWH Retrospective Review

Natasha Kearey, Glen Kennedy, Maree Bransdon and Alanna Geary
Royal Brisbane and Women's Hospital, QLD, Australia

Aim
Retrospective review of patients with Thrombotic Thrombocytopenia Purpura (TTP) post allogenic haematopoietic stem cell transplant.

Method
Retrospective patient chart review. Period of review 2002 – 2007. 266 Allogenic haematopoietic stem cell transplants, 18 developed Thrombotic Thrombocytopenia Purpura, 11 received TPE, 8 other treatments.

Results
Thrombotic Thrombocytopenia Purpura (TTP) in patients who receive allogenic haematopoietic stem cell transplant is a relatively rare complication with a very poor prognosis. The clinical symptoms of post BMT patients seem to parallel those of idiopathic TTP. Often the post BMT transplant patients are treated with therapeutic plasma exchange (TPE) using fresh frozen plasma (FFP) or cryopoor plasma however their treatment remains controversial and largely ineffective given the high mortality rates which remain in this patient cohort.

Idiopathic TTP is understood to be associated with the deficiency of the plasma metalloprotease ADAMTS13. Low levels of this factor cause malfunction of blood clotting, von Willebrand factors which are activated by the endothelial cells and become very large multimers in the circulation unable to be cleaved by ADAMTS13. Microclots form, which consumes the number of circulating platelets and can cause ischaemia of vital organs, low platelets increase patient's risk of haemorrhage and the large strands of von Willebrand factor in circulation destroy red blood cells in their path causing haemolytic anaemia. This can occur from congenital deficiency in ADAMTS13, autoimmune development of antibodies against ADAMTS13, reaction to some drugs or infections. Performing plasma exchange in these cases leads to replacement of the deficient ADAMTS13 and in cases of autoimmune TTP, removes the antibodies against ADAMTS13. Although there have been studies overseas to examine the activity of ADAMTS13 in cases of post allogeneic haematopoietic stem cell transplant TTP, this blood test is currently unavailable in Australia.

Conclusion
Recent studies suggest that development of TTP in post BMT patients is a result of damage to endothelial cells caused by immunosuppressant medications, conditioning regimes, total body irradiation (TBI) and acute graft versus host disease (GVHD). This retrospective review will compare successful treatment of acute GVHD and the resolution of TTP.

Monday 15 October **0830-1000**
Nursing Free Communications 1 **Meeting Room 5**

O41 0930

DC Apheresis for Novel Immunotherapy Strategies

Sonia Hancock[1, 2], Frank Vari[1], Georgina Crosbie[1], Rebecca Prue[1] Jennifer Freeman
[1]*Mater Medical Research Institute (MMRI), Brisbane, Qld, Australia,*
[2]*Apheresis Unit, Mater Health Service (MHS), Brisbane, Qld Australia*

Aim
Develop and optimise a semi automated leukapheresis method to obtain sufficient numbers of blood dendritic cells (BDCs) for use in novel immunotherapy clinical trials.

Method
Between June 2005 and July 2007, 34 healthy, 9 prostate and 4 multiple myeloma donors underwent Auto PBSC collections using a Cobe Spectra machine. Each DC collection processed between 10-15 L of total blood volume. In order for the interface to fully establish itself, the first harvest was not collected before 2L of TBV was processed. Harvest and chase volumes were altered based on the estimated haematocrit content – as judged by the operator using the Spectra Colorgram. A bag count was performed on the end product. DC enriched preparations were produced by immunoselection using either the Miltenyi BDCA-1 kit or the murine CMRF-56 antibody and flow cytometric analysis was performed to determine the number of BDCs ($CMRF56^+$ and $BDCA\text{-}1^+$) obtained.

Result
Currently, the majority of clinical trials in DC immunotherapy use monocytes which require extensive cell culture to produce DCs. Immunoselection of preformed circulating BDCs is a logical source of antigen presenting cells that appear to have a number of advantages both logistical and functional over these “manufactured’ DCs. The average number of DC recovered is 55×10^6 cells using the CMRF56 method and 3.95×10^6 cells using the BDCA-1 kit. Using our optimised apheresis method, we collect sufficient numbers of BDCs for use in novel immunotherapy clinical trials. The procedure is well tolerated by all donor groups with minimal side effects.

Conclusion
Apheresis for novel immunotherapy trials is an emerging subspecialty of haematology. Many patients undergoing this procedure are anxious to maximise the recovery of DCs for vaccine production. These data show that the apheresis method used obtains sufficient yields of $CMRF56^+$ and $BDCA\text{-}1^+$ BDC for use in novel immunotherapy clinical trials.

Monday 15 October **0830-1000**
Nursing Free Communications 1 **Meeting Room 5**

O42 0945

Cellular Therapy for Acute Graft Versus Host Disease – Are Mesenchymal Stem Cells the Panacea?

Debbie Hayes[1], Terry Ventrice[2]
[1] *CNC Bone Marrow Transplant Coordinator, Royal Adelaide Hospital, Adelaide, South Australia, Australia*
[2] *CNC Haematology/Bone Marrow Transplant Unit, Royal Adelaide Hospital, Adelaide, South Australia, Australia*

Allogeneic Bone Marrow (BM) or Peripheral Blood Stem Cell (PBSC) Transplants have become a widely accepted curative treatment for patients with specific haematological malignancies. Graft versus host disease (GvHD) is a major complication of allogeneic transplants contributing to significant morbidity and mortality. It is usually treated with immunosuppressive drugs and high doses of steroids. Treatment for patients who become steroid resistant are a particular challenge. The Royal Adelaide Hospital (RAH) is running a study for patients at risk of developing steroid resistant GvHD. Targeted patients are all patients undergoing a Matched Unrelated Donor Transplant or a high risk transplant (ie mismatched or extended family donors).

Haemopoietic stem cells are found within the bone marrow and are responsible for the production of healthy blood cells. Mesenchymal Stem Cells (MSC) are also found within the bone marrow in very small numbers. MSC's are pluripotent non-haemopoietic precursors that give rise to various connective tissue lineages such as bone, cartilage and adipose tissue. They have also been shown to have immunosuppressive properties and therefore may be useful in the treatment of severe GvHD.

Donors do not need to be HLA identical, have the same blood group as the recipient, or even be related. Donors are screened, educated and consented for the study prior to undergoing a bone marrow biopsy where a 40ml bone marrow aspirate is taken. The bone marrow is used to grow MSC's in the transplant laboratory on site, which takes approximately 6-8 weeks, and once adequate cells have been cultured they are cryopreserved to potentially be infused at a later date. The study is designed to evaluate whether an MSC infusion will help in the treatment of steroid refractory GvHD.

The clinical experience of MSC infusion is limited. There have been some studies and case reports that demonstrate MSC expansion is feasible and infused MSC's do not produce adverse effects. So, are they the panacea?

This discussion will describe MSC's and their application in steroid resistant GvHD. It will outline the current study being undertaken at the RAH including donor identification, the collection and culturing of the MSC's, the infusion process and study results to date. Nursing implications and care of this patient population will also be discussed.

Monday 15 October **0830-1000**
Nursing Free Communications 2 **Meeting Room 6**

O43 0830

Gardeners, Ward persons, Administration, Nurses, Doctors – The Challenge of an Organisational Wide Cytotoxic Awareness Training Program

Leisa Brown, Rachel Edwards, Michael Smith, Maree Bransdon, Alanna Geary
Royal Brisbane and Women's Hospital QLD, Australia

Aim
Develop and implement a sustainable organisational wide Cytotoxic Management Plan

Method
Identified departments where employees may be exposed to cytotoxic drugs and related waste.
Prioritised departments according to risk of exposure to cytotoxic drugs and related waste.
Developed and conducted Cytotoxic Drugs and Related Waste Audits.
Conducted Risk Assessments of areas involved with cytotoxic drugs and/or related waste.
Identified specific training requirements for employees at risk of exposure to cytotoxic drugs and related waste.
Developed a variety of training sessions related to the safe handling of cytotoxic drugs and related waste:
Training sessions based on learning needs of various employee categories.
Conducted training sessions for high priority areas as requested.
Developed a Train the Trainer Program.
Developed a Resource package for areas outside of Cancer Care.
Revised the current Intranet based training package.
Conducted the Train the Trainer Program.
Implemented the Cytotoxic Management Training Program.
Supported implementation process.
Evaluated the Cytotoxic Management Training Program.
Reviewed the Cytotoxic Management Training Program as required.

Results
Reduced risk to patients, staff and visitors of cytotoxic exposure;
Reduced risk to the organisation by complying with the legislative obligations regarding hazardous substances;
Development and implementation of a strategic, organisationally focused cytotoxic training and management plan;
Improved staff awareness and ability to deal with cytotoxic drugs and related waste;
Improved capacity for consistent and coordinated service delivery.

Conclusion
All employees who may be at risk of exposure to Cytotoxic drugs and related waste will receive Cytotoxic awareness training. No employees will carry out work involving cytotoxic drugs and related waste until they have received the appropriate training and instruction.

Monday 15 October **0830-1000**
Nursing Free Communications 2 **Meeting Room 6**

O44 0845

Outpatient Administration of Consolidation Chemotherapy for Acute Myeloid Leukaemia

D Carr, C Milton, C Cairney
Department of Haematology, Newcastle Calvary Mater, Newcastle, NSW, Australia

Aim
There has been an increase in the number patients being treated for Acute Myeloid Leukaemia at the Calvary Mater Newcastle. To accommodate this increase and in response to the demand on inpatient beds within our haematology unit innovative treatment regimes needed to be developed to manage the increased demand safely and efficiently without compromising patient care.

Optimal care requires patients to receive their treatment on schedule. Delays due to lack of inpatient bed availability can compromise this care. One of the treatment protocols developed was the Administration of Consolidation Chemotherapy for Acute Myeloid Leukaemia as an Outpatient.

Method
Outpatient Autologous Stem Cell Transplant have been provided by our Unit over the last 6 years so it was felt that similar suitability criteria and restrictions could be adapted from this protocol.

Patient suitability for the outpatient consolidation regime was discussed on an individual basis at the unit's weekly multidisciplinary meetings. This enabled all members of the multidisciplinary team to have input into the patient's fulfillment of the required criteria. The availability of this option was then discussed with the patient in order for the patient to be allowed the opportunity to accept or decline being part of the program.

In order for the program to be successful pharmacy needed to be able to adapt the standard consolidation regimes for AML to suit the outpatient environment but also to not impact on the efficacy of the therapy required.

Another major factor to the success of this program was the appropriate placement of a Central Venous Access Device to ensure the safety and efficacy of chemotherapy delivery.

Results
The success of this pilot study has been measured by evaluating set outcome measures retrospectively and by measuring patient satisfaction outcomes.

Conclusion
The Haematology Unit at the Calvary Mater Newcastle has successfully implemented the administration of outpatient consolidation chemotherapy regimes for the last twelve months and plans on continuing this service on an ongoing basis.

Monday 15 October **0830-1000**
Nursing Free Communications 2 **Meeting Room 6**

O45 0900

Developing a Febrile Neutropenia Protocol for the Emergency Department

Allan Hayward
Clinical Operations Manager, Institute of Medical and Veterinary Science/Royal Adelaide Hospital, Adelaide, South Australia, Australia

Febrile Neutropenia is a very serious and potentially life threatening complication following chemotherapy administration. Increased pressure on inpatient beds has meant a move towards outpatient chemotherapy administration or early discharge following chemotherapy. Patients are instructed to return to the hospital Emergency Department (ED) should they become febrile or unwell.

There have been a number of strategies in place to expedite the patient journey through ED including supplying high risk patients with a covering letter, medication and investigation orders, as well as an ED protocol that had not been revisited in a number of years. It was found that these measures did not correlate with an optimal patient experience on their return to the hospital's ED, reflected in long waiting times to be seen, to the commencement of antibiotics and the commencement of investigations. As a result, the management of these patients in the ED became a high priority.

A number of departments have been working together to develop a more comprehensive protocol for the management of the febrile neutropenic patient presenting to the ED. This has involved extensive consultation with haematology, emergency, infectious diseases and the safety and quality unit.

The result has been a single page protocol that outlines the immediate care needs of the patient presenting to ED with febrile neutropenia. The protocol includes a flow chart that helps staff identify patients at risk of febrile neutropenia, outlines the investigations and antibiotics that should be initiated along with suggested timelines, contact details for haematology/oncology medical staff as well as considerations for ICU admission and home ward admission.

This new and revised protocol was launched in June of this year, accompanied by an introductory inservice to ED medical and nursing staff. This presentation will discuss the protocol and the results of clinical audits currently being undertaken; both before the introduction of the revised protocol and following its implementation.

Monday 15 October **0830-1000**
Nursing Free Communications 2 **Meeting Room 6**

O46 **0915**

A Review of Central Line Data

David Collins
BMT Network NSW, Sydney, Australia

Patients undergoing treatment for haematological malignancies or bone marrow transplantation will require some form of central venous access. However, for many of these patients infection can be a problem. These infections can be serious and result in the loss of the line. A literature review was undertaken, but very little was found with regard to line infections in immunocompromised patients. Data of patients requiring central venous access have been collected in a haematology unit and reviewed. This paper will review these results looking for common links that may demonstrate factors that predispose patients to line infections and make recommendations for further research.

Monday 15 October **0830-1000**
Nursing Free Communications 2 **Meeting Room 6**

O47 **0930**

Intra-ventricular Treatment: Experiences at RBWH

Carmen Worthey, Maree Bransdon, Ron Middleton, Glen Kennedy, Alanna Geary

Aim
To evaluate the effectiveness of Rituximab, Methotrexate and Cytarabine administered intra-ventricularly by nursing staff.

Method
Nurses, when determined competent, administer intra-ventricular Rituximab, Methotrexate and Cytarabine.

Recent literature expresses the opinion that intravenous administration of Rituximab has limited penetration into the leptomeningeal space. Papers focusing on recent trials have concluded that the administration of intra-ventricular Rituximab together with Methotrexate and Cytarabine for Diffuse Large B-cell Lymphoma (DLBCL) with CNS involvement, recurrent CNS lymphoma, mantle cell lymphoma, and anaplastic lymphoma with CNS involvement is effective. [1]

This method is being administered by nursing staff. Patients with Primary CNS Lymphoma (PCNSL) and Post Transplant Lymphoproliferative Disorder (PTLD) received this treatment option using:

- an Ommaya reservoir;
- a 25g butterfly needle;
- Betadine;
- a calculation of CSF sampling, discard and flush;
- an administration time of 1ml/minute when administering Methotrexate and Cytarabine; and
- an administration time of 10 – 15 minutes when administering 25mg of Rituximab.

Results
Nurses have been successfully administering Rituximab, Methotrexate and Cytarabine at RBWH without any adverse incidents or device infections. The results of the consulted papers are reflective of experiences at RBWH with the administration of Rituximab. The Giovanni and Rubenstein study reported no major dose limiting side effects. The Wang and Villela studies reported only minor side effects (grade I-II) but all were resolved within 10 to 20 minutes. Further, all papers reported no side effects with the administration of Methotrexate and Cytarabine intra-ventricularly without Rituximab.

Conclusion
Nursing staff at RBWH are covering new ground by administering Rituximab, Methotrexate and Cytarabine intra-ventricularly. The absence of device infections indicates training procedures for nurses using this method are effective. Intra-ventricular Rituximab together with Methotrexate and Cytarabine is effective in the treatment of Lymphoma's with CNS involvement, is well tolerated and has seen a significant extension in survival.

Reference
Rubenstein (2007), Villela (2006), Giovanni (2007) and Wang (2006)

Monday 15 October **0830-1000**
Nursing Free Communications 2 **Meeting Room 6**

O48 0945

A Single Centre Experience of Cytosine Toxicity

Patricia Ryan
Haematology, Liverpool Hospital, NSW

This paper discusses a single centre experience of acute cerebellar toxicity with ataxia and dysarthria in 3 patients receiving high-dose cytosine arabinoside therapy.

High dose cytosine arabinoside has been a useful addition to a number of regimens used in treating a number of haematological malignancies including Acute Leukaemias and high grade lymphomas. In addition to the usual toxicities associated with antimetabolites, neurotoxicity, mainly in the form of cerebellar dysfunction, develops in a significant proportion of patients receiving high-dose cytosine arabinoside (HDara-C). Initial toxicity symptoms related to mucosal breakdown especially of conjunctival and gastrointestinal surfaces, but more extensive trials with high-dose intermittent administration schedules identified neurotoxicity as a serious potential side effect of this therapy. The problem has been noted to be dose-related, appearing at total doses over 18 g/m^2 per course. The incidence has been estimated to range from 0% to a maximum of 16.7% in various studies [1,2].

Although acute cerebellar toxicity with ataxia and dysarthria is a well-known side effect during high-dose cytosine arabinoside therapy. Dose, age, previous neurological disorders, age [3], hepatic dysfunction [4], and renal insufficiency have been inconsistently reported as risk factors [5]. This paper discusses a single centre experience of 3 cases of acute cerebeller toxicity in patients undergoing treatment for haematological malignancy.

References

1 Winkelman MD, Hines JD. Cerebellar degeneration caused by high-dose cytosine arabinoside: A clinicopathological study. *Ann Neurol* 1983; 14:520-527.
2 Early AP, Preisler HD, Higby DJ *et a/.* High-dose cytosine arabinoside: Clinical response to therapy in acute leukaemia. *Med Pediatr Oncol* 1982; (Supp1)10:239-250.
3 The neurotoxicity of high-dose cytosine arabinoside is age-related. D Gottlieb, K Bradstock, J Koutts, T Robertson, C … - Cancer, 1987
4 Neurotoxicity associated with systemic high-dose cytosine arabinoside. S Nand, HL Messmore Jr, R Patel, SG Fisher, RI - J Clin Oncol, 1986
5 Cerebellar toxicity during cytarabine therapy associated with renal insufficiency. -H Hasle - Cancer Chemotherapy and Pharmacology, 1990

Monday 15 October **1030-1100**
HSANZ Symposium: Immunotherapy **Arena 1B**

1030

Lymphoma Vaccines

Ronald Levy
Division of Oncology, Stanford University Medical Center, USA

Two different strategies of therapeutic vaccination against lymphoma will be presented: 1. the defined antigen approach with custom made idiotypes derived from each patient's own tumor and 2. Forced antigen presentation by injecting CpG, an immune activator into a single tumor site after apoptosis is induced by low dose radiotherapy.

Each of these strategies has been tested extensively in preclinical animal models and each is now the subject of clinical trials in humans. Three separate phase III clinical trials of idiotype vaccination are nearing completion and they will be compared and contrasted. The final results will not be available at the time of the meeting but they are eagerly anticipated by the start of the New Year. Retrospective updating of earlier phase II trials with long term survival outcomes indicates that antibody immune responses correlate strongly and independently with survival. One pilot clinical trial of CpG intratumoral injection has begun in patients with relapsed low grade B cell lymphoma and mycosis fungoides and interim results will be presented

Monday 15 October **1030-1100**
HSANZ Symposium: Immunotherapy **Arena 1B**

1100

Challenges for Immunotherapy

Ian Frazer
Diamantina Institute for Cancer, Immunology and Metabolic Medicine, Brisbane, Qld, Australia

Specific Immunoprophylaxis against infection has been one of the successes of medical research. However, we have yet to develop specific immunotherapy to alter the course of chronic infection, autoimmunity or cancer. A vaccine to prevent cervical cancer required only the recognition that cervical cancer is a consequence of persisting infection with human papillomavirus, and a technology for production of a virus mimic to induce neutralising antibody. In contrast, specific immunotherapy will require appropriate cellular immune responses, and we are unsure what is required to produce these. An animal model of immunotherapy for cervical cancer will be discussed in which it can be demonstrated that defining the immunogen and the cellular response may be less important than modulating the local environment in which the immune response is generated. The conclusion is that successful immunotherapy may require a system of measures to reset the immune system including neutralisation of inhibitors of LL-1 and IL10.

Monday 15 October **1030-1130**
ANZSBT Symposium: Transfusion Competencies **Central Room A**

1030

Training in Transfusion – UK Perspective

Geoff Daniels
Bristol Institute for Transfusion Sciences, NHSBT, Bristol, UK and the British Blood Transfusion Society

Career structures, education, and training for healthcare professionals are currently in flux in the UK. A career framework for healthcare scientists has been developed by the government through an external agency, Skills for Health (SfH). The framework consists of 9 levels, ranging from level 1 (posts requiring very little formal education or previous knowledge) to level 9 (very senior scientists and consultant healthcare scientists directors). It is now important that the appropriate education and training is available to enable healthcare scientists to obtain state registration with the Health Professions Council (HPC) and to progress up through the career framework.

Professional bodies representing UK healthcare scientists in the transfusion field include the British Blood Transfusion Society (BBTS), the Institute for Biomedical Sciences (IBMS), the British Society for Histocompatibility and Immunogenetics (BSHI), and the Royal College of Pathologists (RCPath). These organisations together with higher education facilities provide educational and training courses and qualifications. These include many coterminous BSc degrees with transfusion content, leading to state registration as a biomedical scientist Two MSc courses on transfusion and on transfusion and transplantation sciences, and many more MSc courses with a transfusion element, are available. The BBTS currently provides a Specialist Certificate in Transfusion, and BBTS Specialist Certificates in Transfusion Science Practice, for nurses and transfusion practitioners, and in Cell and Tissue Transplantation Sciences, will be available in 2008. Other relevant Certificates and Diplomas are provided by the IBMS. The UK national blood services also provide training courses and there are a number of e-learning modules available. Unfortunately the RCPath does not currently provide a membership examination for Clinical Scientists that contains a substantial transfusion content. Continuing professional development (CPD), based on reflective learning rather than the accumulation of points, is essential for maintenance of HPC registration.

Monday 15 October **1030-1130**
ANZSBT Symposium: Transfusion Competencies **Central Room A**

1100

e-Learning and Transfusion Medicine

David Peterson, Kathryn Robinson, Trudi Verrall, Bev Quested, Ben Saxon
BloodSafe: Transfusion Safety and Quality Improvement Program, Department of Health and Australian Red Cross Blood Service, South Australia

Audits of clinical transfusion practice [1] have consistently demonstrated deficiencies in knowledge and practice that impact on patient safety and in some cases result in death. Improvement needs to be driven by a multifaceted approach that includes robust systems and appropriate staff knowledge combined with mechanisms to ensure compliance.

A mechanism for improvement has been to use hospital accreditation processes, with the ANZSBT contributing new criteria to the ACHS EQuIP (4^{th} Edition) process that specifically address transfusion practice. One of the specific criteria is for knowledge credentialing of staff involved in the transfusion process. However, large numbers of hospital staff, shiftwork, varying levels of background knowledge and limited resources create significant staff education challenges. Needs analysis defined an on-line learning tool as being a suitable mechanism to assist hospitals to meet these requirements. Funding was provided under the BloodSafe program by the South Australian Department of Health to develop an on-line learning tool.

An effective e-learning tool must be engaging and replicate an authentic learning environment. This requires knowledge of on-line learning best-practice, learner profiles, learning styles and the learning environment, as well as consideration of interface design, motivational tools and knowledge retention strategies. A multimedia rich program utilising video, audio, animations, and case studies (combined with didactic instruction) is used to create an authentic learning environment. A flexible learning pathway gives learners control over learning sequence, content viewed and assessment. Learner demographics, progress and assessment tools are stored in an SQL database.

Following implementation and evaluation at a single hospital site this tool is available for use by other hospitals and healthcare institutions and professionals. Further development is envisaged to provide additional modules offering advanced content and/or a broader audience base.

1 Published and unpublished data from BloodSafe (2003 – 2006) and a number of other groups

Monday 15 October **1030-1130**
ASTH Symposium: Haemostasis **Central Room C**

1030

Do We Understand Type 1 von Willebrand Disease?

Simon A Brown
Queensland Blood Management Program, Queensland Health; Department of Haematology and Children's Haemophilia Centre, QHPS Royal Brisbane and Women's Hospital, and Royal Children's Hospital, Brisbane, Queensland.

It is over 80 years since von Willebrand disease (VWD), the commonest of the inherited bleeding disorders, was first described. Despite the characterisation of von Willebrand factor (VWF) and cloning of the *VWF* gene, the commonest subtype (type 1) of VWD continues to cause clinicians and scientists problems with respect to its diagnosis and pathogenesis. Type 1 VWD is a partial quantitative deficiency of VWF (pqdVWF). In the 1994 classification of VWD, all VWD was assumed to result from a mutation in the *VWF* gene. A large European study was initiated to delineate the nature of the molecular defect in type 1 VWD with the hope of providing a definitive diagnostic test. Together with two other studies from Canada and the UK it is evident that many milder cases of type 1 VWD do not result from mutations in the *VWF* gene. This has lead to a reassessment the pathogenesis of a pqdVWF. One potential pathogenetic mechanism for a pqdVWF is through increased clearance of VWF. Since the reporting of increased clearance of VWF in patients with type 1 VWD (Brown et al. *J Thromb Haemost* 2003) there have been ongoing studies to clarify the role of increased clearance of VWF in pqdVWF and to understand the mechanisms of VWF clearance. Unfortunately, the mechanism of VWF clearance remains poorly understood. Further studies in a cohort of type 1 VWD patients have investigated the factors that may influence VWF clearance, including the ADAMTS 13 proteolytic pathway, ABO blood group and molecular studies. Although the pathogenesis of all cases of type 1 VWD remains unresolved, it is evident that type 1 VWD is not a single pathogenic entity and in some cases may represent a complex genetic disorder.

Monday 15 October **1030-1130**
ASTH Symposium: Haemostasis **Central Room C**

1100

New and Revisited Assays to Assess Haemostasis: Correlation with the Clinical Phenotype

Claude Negrier
Hôpital Edouard Herriot, University of Lyon, France

The coagulation process involves cellular surfaces on which the haemostatic proteins bind and interact together. In the case of vascular injury, tissue factor and factor VII/ VIIa interact at the surface of tissue factor–bearing cells to produce extrinsic factor Xase through activation of both factor IX and factor X. Complexed with factor Va, factor Xa activates small amounts of thrombin that are insufficient to cleave fibrinogen. This thrombin can initiate an amplification mechanism by activating platelets, causing release of factor V from storage granules, then activating factor V, releasing factor VIII from von Willebrand factor and cleaving factor VIII, and activating factor XI. The formation of factor Xa/factor Va complex results in a burst of thrombin generation, which is able to cleave fibrinogen and form a fibrin clot. In haemophilia, initiation of the coagulation proceeds normally, but there is a failure of factor X activation on the platelet surface, leading to a dramatic decrease in thrombin generation and ineffective clot formation. Therefore, thrombin generation or fibrin formation could represent useful surrogate markers to assess the bleeding or thrombotic tendency, the efficacy of therapeutic agents *in vitro* and the achievement of haemostasis *ex vivo*. Real-time continuous whole blood clot formation can be recorded by a thromboelastography (TEG) analyser, and it was demonstrated that the velocity profiles from patients with haemophilia were considerably different from reference values. The kinetics of thrombin generation can also be monitored using a specific software and a fluorimeter. However, significant inter-individual variations were also detected. These assays offer the potential to choose a therapeutic product and to tailor the dose according to the haemostatic response to varying doses tested prior to *in vivo* administration. Despite promising results, the correlation with *in vivo* clinical response needs further investigation.

Dr Claude Negrier sponsored by CSL Bioplasma

Monday 15 October **1030-1130**
Nursing Free Communications 3 **Meeting Room 5**

O49 1030
Establishing a Collaborative Transfusion Clinic

Sharron Ellis
Christchurch Hospital; Christchurch; New Zealand

Introduction
A rapid growth in patient numbers, creating stress and frustration for medical staff already working at maximum capacity, meant that urgent changes to some out-patient clinics was required.

Problems
One issue that was identified was a lack of time for medical staff to comprehensively assess patients who require frequent blood transfusions in clinics that only had fifteen minute allotments. It was felt that the needs of these patients would be better met in a nurse-led clinic where there was more time available.

Solving the problems
Therefore it was proposed that a nurse-led clinic would be established in the outpatient setting to address this need. It would provide a service to these patients that was holistic, maintained patient safety, improved patient outcomes and allowed senior nurses professional autonomy and the opportunity to broaden their scope of practice. The nurses would receive education in health assessment to master's level.

A proposal was written outlining the requirements to establish the clinic. The Director of Haematology approved the proposal after funding issues were addressed. Currently there is a nurse being educated to Masters Level for health assessment of the adult. This nurse will begin assessing patients in a clinic by the end of 2007.

Conclusion
This is an exciting time for the Haematology Service. There have been many challenges and delays. The process has taken three years to get to the point where the clinic is becoming a reality. It is also a huge opportunity for other nurses in the service to take up the challenge to work in this clinic so that more patients can be seen. It is agreed by all those involved in this proposal that the outcomes will be a safer, more holistic service for patients.

Monday 15 October **1030-1130**
Nursing Free Communications 3 **Meeting Room 5**

O50 1045

Management of the Febrile BMT and Haematology Patient in the Emergency Department- A Management Tool for Nurses

Bettina Clark
Bone Marrow Transplant Coordinator, The Royal Melbourne Hospital

Patients who have undergone treatment for haematological malignancies, or a Bone Marrow Transplant (BMT) are highly immunosuppressed and are therefore extremely susceptible to infection. Many of these patients may present to the Accident and Emergency Department (A&E) with a fever. Fever due to infection in these patients can rapidly progress to septic shock and death, if appropriate, prompt, and effective treatments are not commenced immediately on presentation to hospital.

As there are numerous potential sources of infection in this group of patients a thorough septic workup and treatment is essential. On reflection of recent practice, we found that often workups were incomplete, samples labelled inappropriately, and completion of a workup was reliant on the patient being neutropenic regardless of risk. A management tool was developed for nursing staff that specifically outlined the management of febrile haematology and BMT patients on presentation to A&E. This tool provided the nursing staff with an increased knowledge of the requirements for a complete septic workup, and awareness of the need for prompt treatment in this high-risk patient group.

The implementation of this tool has decreased staff anxieties and increased their knowledge when treating these patients. Prompt assessment and intervention for these patients in A&E, has decreased repeating of tests and improved outcomes for these patients. In turn, patients report feeling less anxious as they feel reassured that they will be appropriately treated if they should need to present to A&E.

Monday 15 October **1030-1130**
Nursing Free Communications 3 **Meeting Room 5**

O51 1100

CI-SCaT: An Adjunct to Streamlining the Patient Journey

A Booth[1], S Rushton[1], J Bichel-Findlay[1], R Ward[2], S Sinclair[3]
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[2] *Department of Medicine, Prince of Wales Hospital, Sydney*
[3] *Cancer Services and Education Division, Cancer Institute (NSW), Sydney*

The delivery of best practice nursing care to cancer patients requires a sound understanding of the contemporary literature, key evidence and internationally accepted standards. In an environment of rapidly changing and increasingly complex treatments, compounded by a less experienced/specialised and time-poor workforce, haematology nurses are finding it more difficult to achieve this in a timely manner.

The Cancer Institute NSW Standard Cancer Treatment program (CI-SCaT) content team, in conjunction with nurse clinicians from NSW and interstate, identify clinically relevant protocols for inclusion on this web-based resource. The CI-SCaT team develops the protocols based on the latest evidence blended with current practice. After discussion with, and approval by, the nurse clinicians, the completed protocols are uploaded to the website. Protocols provided are safe, evidence-based, freely available online, provide expert guidance and may negate the necessity for each individual facility having to write, review and update protocols.

CI-SCaT has now been available for two years, over which time the patterns of uptake and acceptance have changed. There were a number of early change champions who embraced the resource from the start as it was obvious how it would aid local practice. For others, CI-SCaT was, and is perceived as, a challenge to the autonomy of individual practice, but more recently it appears there is an increasing recognition of the potential benefits as use of the website increases.

Work is underway on an independent evaluation of CI-SCaT's integration into the clinical environment, development of a more flexible and functional website, and finalisation of the strategic plan up to 2010, all of which have been guided by extensive clinician input.

The increasingly widespread acceptance of CI-SCaT as a reliable source of nursing protocols blending evidence with practical realities will, it is hoped, lead to a more streamlined patient journey and, therefore, improved patient outcomes.

Monday 15 October **1030-1130**
Nursing Free Communications 3 **Meeting Room 5**

O52 1115

Living Longer, Living Differently? The Changing Experience of Living with Multiple Myeloma

Moira Stephens
The University of Sydney, Sydney, NSW, Australia

Aim
To review research into the experience of surviving multiple myeloma (MM).

Background
People are now living longer with MM, but little is known about how they manage this experience.

Methods
Databases were searched for relevant literature published between 1960 and 2007. Experts were asked for key references and ongoing studies. Internet searches were used to locate "Blogs" and virtual (online) support groups. Actual support groups were observed.

Results
Among those diagnosed with MM, median duration of survival increased from several months in the 1960s (Osgood, 1960) to 3 years in the 1990s. Median survival is now 7.5 years in patients <50 years and 5.7 years in older patients, and 43% of younger patients and 29% of older patients survive 10 years (Ludwig et al in press). Many patients survive 10-15 years. Because MM remains incurable, the main goals of treatment are to control disease, secure remission and maximise quality of life. Most patients die from their disease despite onerous and often complex regimens involving multi-disciplinary support and different treatment modalities. Although it is widely acknowledged that living with and dying from MM is resource intensive and often a painful and difficult journey, there is scant research into the experiences of people with MM and none on those living with MM in a chronic state of relapse.

As treatments change and survival is extended, the experience of living with MM also changes. Survivors are sharing knowledge and experience outside of clinical settings in support groups (actual and virtual) and in "MM blogs". The latter reveal how people are managing the experience of living longer with MM.

Conclusions
Increased duration of survival has implications for service delivery and future research into MM. Understanding how people manage the experience of living longer with MM can help tailor services to patient's needs.

Reference
OSGOOD, EE (1960) The Survival Time of Patients with Plasmocytic Myeloma. *Cancer Chemotherapy Reports*, 1-10.

Monday 15 October **1130-1230**
HSANZ: Carl de Gruchy Oration **Arena 1B**

Lessons from the Clinic: Are There Known Knowns?

Michael C Berndt
Monash University, Melbourne, Victoria, Australia

Decades before the development of genetically-modified mice, analysis of patients with bleeding abnormalities led to our current understanding of coagulation and haemostasis. Analysis of patients with von Willebrand's Disease, Bernard-Soulier Syndrome and Glanzmann's Thrombasthenia defined the importance of von Willebrand Factor, the GPIb-IX-V complex and GPIIbIIIa (□IIb□3), respectively, in platelet adhesion and aggregation. Similarly, in the late 1980s, the role of GPVI as the key collagen receptor on platelets was initially determined through analysis of rare GPVI-deficient patients. Whilst more recent studies with genetically-modified mice have recapitulated these earlier clinical observations in haemostasis, their analysis has identified an unexpected role for some of these proteins in thrombosis. In particular, Factor XI and Factor XII deficient mice, as for human patients deficient in these two Factors, either have a normal bleeding time or only a very mild bleeding diathesis. Platelets from these mice however have a profound defect in their capacity to form occlusive thrombus in a number of distinct arterial thrombosis models. This can be understood if the role of platelets and coagulation are not considered in isolation but as intimately-linked processes in thrombus development. Recent data will be presented demonstrating a critical role for the intrinsic pathway of coagulation in mediating platelet-dependent thrombin formation.

Monday 15 October **1330-1500**
HSANZ ASTH Presidential Symposium **Arena 1B**

O53 1330

Deregulation of a Cytokine Receptor Phospho-Ser/Phospho-Tyr Binary Switch in Acute Myeloid Leukemia

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[4] Department of Haematology, Institute of Medical and Veterinary Science, Hanson Institute, Adelaide, SA, Australia 5000

Aim
Constitutive activation of cytokine signaling pathways that lead to deregulated hemopoietic cell proliferation *and* survival underpin cellular transformation and leukemogenesis. We have previously identified a conserved phospho-Tyr/phospho-Ser binary switch in the cytoplasmic tail of GM-CSF receptor which is composed of Tyr577 and Ser585 that allows the independent regulation of hemopoietic cell proliferation and survival. We examined the status of this receptor in acute myeloid leukaemia (AML) blasts from pre-treated patients. We also wished to investigate genes that are regulated by Ser585 in AML and obtain data on the survival response of AML blasts in presence of serine/threonine kinase inhibitors compared to tyrosine kinase inhibitors.

Method
Blasts from consenting patients were examined for receptor phosphorylation status in response to cytokine using phosphospecific antibodies to Ser585 and Tyr577. Cytokine-mediated survival examined by annexin V staining of blasts in presence of selective kinase inhibitors. Microarray analysis for Ser585 dependent genes was performed using in CTL-EN cells expressing wild-type and mSer585Gly mutant GM-CSF receptors. Bayesian analysis and moderated t-statistics were used to identify Ser585-regulated genes. Mann-Whitney *U* test was used to confirm differences in gene expression between AML patients and normal monocyte controls.

Results
Ser585 is constitutively phosphorylated in AML blasts (18/21 patients). In contrast, phosphorylation of Tyr577 remains cytokine-dependent (20/21). Pharmacological blockade of protein kinase A (PKA) reduced constitutive Ser585 phosphorylation and autonomous survival of AML blasts from some patients whereas tyrosine kinase inhibitors had no effect on either Ser585 phosphorylation or cell survival. We have identified a panel of Ser585-regulated genes from which we have validated a "four-gene Ser585-signature" that is deregulated in AML blasts exhibiting constitutive Ser585 phosphorylation.

Conclusion
Our results link a distinct form of constitutive cytokine receptor activation to cell survival and support the notion that deregulated survival signals allow the persistence of minimal residual disease and increase the likelihood of relapse.

Monday 15 October **1330-1500**
HSANZ ASTH Presidential Symposium **Arena 1B**

O54 1345

Reduced Fibrinolysis and Increased Fibrin Generation is seen in Patients with Thrombotic Complications of Pregnancy Compared with Normal Pregnancy

Monika Mukerji[1], Ninfa Rojas[1], Jocelyn Sedgely[2], Marie-Christine Morel-Kopp[1,2], Christopher M Ward [1,2] and Jennifer L Curnow[1,2]
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[2] Department of Haematology and Transfusion Medicine, Royal North Shore Hospital, Sydney, Australia

Pregnancy is a physiological hypercoagulable state which may be complicated by venous thromboembolism (VTE) or miscarriage. We postulated that the OHP, a global coagulation assay, would be hypercoagulable in normal pregnancy when compared with the non pregnant state and that the assay parameters would indicate greater hypercoagulability in individuals with increased risk of VTE or miscarriage.

Patients with healthy pregnancy were tested to determine an OHP reference range for pregnancy, for each trimester. For the OHP assay, thrombin (0.03 U/ml) was used to trigger fibrin generation in platelet poor plasma (PPP) with rt-PA (300 ng/ml) added to initiate fibrinolysis. OHP results were compared with reference intervals determined in a reference population of 200 healthy non-pregnant Australians. OHP parameters were hypercoagulable with increased fibrin generation and reduced fibrinolysis in second and third trimesters of healthy pregnancy when compared with a non-pregnant population.

15 pregnant patients referred to clotting clinic for recurrent miscarriage or VTE, some in association with thrombophilias, were also examined. Patients had recurrent miscarriage (n=4), previous VTE (n=8) or thrombophilia without a prior event (n=3). Thrombophilias included antithrombin deficiency (n=3), protein S deficiency (n=4), FVL heterozygous (n=2) and antiphospholipid syndrome (n=2). In this group, when tested prior to clexane use, fibrinolysis was further reduced and fibrin generation increased compared with healthy pregnancy. Commencement of clexane altered fibrin generation such that mean values were similar to healthy pregnancy, however fibrinolysis remained reduced in the patient group.

OHP assay findings are consistent with the physiological hypercoagulable state associated with normal pregnancy. OHP assays in pregnant patients with thrombotic complications, recurrent miscarriage and/or thrombophilias indicate further hypercoagulability compared with healthy pregnancy. In most cases, fibrinolysis remains abnormal despite the introduction of clexane.

Monday 15 October **1330-1500**
HSANZ ASTH Presidential Symposium **Arena 1B**

O55 1400

Combining RAD001 (Everolimus) with DNA Damaging Agents as a Therapeutic Strategy Against Precursor-B Acute Lymphoblastic Leukemia

Philip Saunders, Linda Bendall, Kenneth Bradstock
Westmead Institute for Cancer Research, Westmead Millennium Institute, Sydney, NSW, Australia

The past twenty years has produced no significant new agents for the treatment of Precursor-B Acute Lymphoblastic Leukemia (Pre-B ALL). Mammalian target of rapamycin (mTOR) inhibitors have shown potential as novel therapeutic agents with efficacy against a wide range of tumors. We performed in vitro studies to assess the efficacy of RAD001 (Everolimus; Novartis) against Pre B ALL. RAD001 inhibited tumor proliferation in a dose dependent fashion (IC_{50} 1-2 μM) in Pre-B ALL cell lines, with a marked increase in the proportion of cells in $G_{0/1}$ cell cycle arrest. Dose escalation induced cell death in both cell lines and untreated primary cases (LD_{50} 4-16μM).

While RAD001 has potential as a single agent, combining escalated doses of RAD001 with DNA damaging agents, as induction or salvage therapy is most likely to maximize the potential of this novel agent. We observed caspase dependent synergistic killing of Pre-B ALL blasts combining RAD001 (8-16μM) with sub-LD_{50} doses of Vincristine, Doxorubicin, Etoposide and ionizing radiation at 24 hrs in vitro. Examination of signal protein phosphorylation by intracellular flow cytometry revealed activation of c-jun, a key negative regulator of the p53 and p21 promoter region. In keeping with this finding we observed blockade of p21 induction by DNA damaging agents when combined with RAD001 at 16μM. Analysis of p53 expression suggests p21 is suppressed as a result of c-jun activation and co-suppression of the p53 promoter. We plan further studies to test the hypothesis that synergistic killing is due to suppression of p21 induction by DNA damaging agents with subsequent failure of DNA repair and promotion of apoptosis via a p53 independent pathway.

Monday 15 October **1330-1500**
HSANZ ASTH Presidential Symposium **Arena 1B**

O56 1415

The Bcl-2 Protein Family is Essential for Platelet Survival *in vivo*

KD Mason [1,2], MR Carpinelli [1], JI Fletcher [1], JE Collinge[1], AA Hilton[1], D Metcalf [1], AW Roberts[1,2], BT Kile[1], DCS Huang [1]
[1] *The Walter and Eliza Hall Institute of Medical Research*
[2] *The Royal Melbourne Hospital*

Aims
The steady state peripheral platelet count reflects the balance between platelet production and destruction. The latter occurs when platelets have reached the ends of their life-span. The precise factor(s) controlling the life-span of the platelet are not clearly understood. The observation that ABT-737, a small molecule antagonist of pro-survival Bcl-2 proteins, triggers acute thrombocytopenia led us to investigate if the apoptotic machinery may play a role in regulating platelet life-span.

Methods
We assessed the number, survival and life-span of platelets in mice following pharmacological inhibition or genetic manipulation of the Bcl-2 pro-survival and pro-apoptotic proteins. Protein half-lives were quantified following inhibition of protein synthesis and caspase inhibitors were utilised to evaluate if cell death proceeded by apoptosis. To ascertain if platelet half-life was platelet intrinsic, reciprocal adoptive transfer was performed. The results were confirmed in human platelets.

Results
We discovered that the balance between pro-survival Bcl-x_L and pro-apoptotic Bak controls platelet life-span. *In vivo* platelet survival, hence the number of circulating platelets, is lowered in mice with reduced Bcl-x_L. These changes are platelet intrinsic. *Ex vivo,* both mouse and human platelets die by the process of apoptosis, which is delayed by caspase inhibition. Downstream of Bcl-x_L, removal of the pro-apoptotic protein Bak corrects these defects.

Conclusions
The antagonistic balance between Bcl-x_L and Bak constitutes an intrinsic molecular clock that determines how long a platelet survives. These findings have profound implications for our understanding of platelet biology, and for the diagnosis and treatment of disorders that affect platelet numbers and function. Disorders of platelet number may be a result from inherited or acquired mutations in *Bcl-x* or *Bak,* and modulation of this pathway could potentially be utilized to manipulate platelet numbers for therapeutic gain. Additionally, the ability to prolong platelet survival *ex vivo* could revolutionise platelet transfusion therapy.

Monday 15 October **1330-1500**
HSANZ ASTH Presidential Symposium **Arena 1B**

O57 1430

Pre-transplant FDG-PET Scanning Following Salvage Chemotherapy for Patients with Relapsed Diffuse Large B cell Lymphoma (DLBCL) Predicts Long-term Outcome

Michael Dickinson[1], Rosemary Hoyt[2], Andrew Roberts[2], Andrew Grigg[2], John F. Seymour[1], Miles Prince[1], Jeff Szer[2], David Ritchie[1, 2]

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[2] *Department of Clinical Haematology, Royal Melbourne Hospital, Parkville, VIC*

Introduction

FDG-PET imaging is a powerful predictor of both positive and negative outcome of initial therapy for DLBCL. Existing data on the utility of PET to predict outcome of ASCT have included a range of histological subtypes, limiting its direct applicability to the setting of DLBCL.

Methods

Data from 78 consecutive patients undergoing ASCT for treatment of first relapse of de-novo DLBCL at RMH or PMCC were analysed. We compared the clinical characteristics, event free survival (EFS) and overall survival (OS) of both the whole cohort and those with pre-ASCT PET imaging.

Results

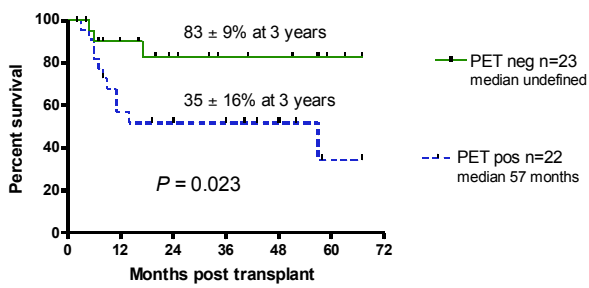
For all 78 patients (median age 52y, 56% male, median aaIPI=1) the median follow up was 26months (2-125), OS =64% and EFS=51% at three years.

Pre-ASCT PET was performed in 45 patients. There were no significant differences in the clinical characteristics, IPI at relapse, salvage regimens used or pre-ASCT conditioning in the PET-positive vs PET-negative patients (Table). 3 year OS and EFS for PET negative (n=23) or PET positive (n=22) patients were 83% vs 35% (p=0.02) and 85% and 36% respectively (p=0.04). Post-ASCT outcome was also favourably influenced by the use of rituximab containing salvage therapy and response to salvage therapy.

Conclusions

Failure to achieve a PET negative remission to salvage chemotherapy in relapsed DLBCL predicts for a poor outcome from ASCT, necessitating the development of novel salvage therapies in this group. Conversely an excellent outcome can be anticipated from ASCT when undertaken in those who have achieved a PET negative remission from salvage.

Overall survival of Autografts with PET positive vs PET negative disease pre ASCT



HR 3.89
95% CI [1.19,9.92]

Monday 15 October **1330-1500**
HSANZ ASTH Presidential Symposium **Arena 1B**

O58 1445

Identification of a Unique Platelet Contractile Mechanism Regulating Thrombus Stability

Simone Schoenwaelder, Akiko Ono, Sarah Hsiao and Shaun Jackson
Australian Centre for Blood Diseases, Monash University, Melbourne, Victoria Australia

Aim
To gain insight into the mechanisms regulating thrombus formation and stability under flow.

Method
Thrombus stability was examined using a well defined *in vitro* flow-based thrombosis assay, independent of blood coagulation. 3D analysis of thrombus volume was performed using confocal microscopy.

Results
Real-time analysis of thrombi forming on a collagen matrix, but not on a VWF or fibrinogen matrix, revealed a distinct stage of thrombus development, which we have termed thrombus contraction. Thrombus contraction was apparent during the formation of large platelet aggregates on a collagen substrate and typically occurred over the first 5 minutes of thrombus development. Real-time 3-D analysis of forming thrombi by confocal microscopy revealed that thrombus contraction was associated with a 30-40% reduction in thrombus size (n=12). Thrombus retraction was shear-dependent, with the rate of retraction increasing as a function of increasing blood flow rates. This retraction was accompanied by consolidation of the thrombus core, suggesting that thrombus contraction may play a role in promoting thrombus stability. Inhibiting the platelet contractile mechanism by pretreating platelets with the specific myosin IIa inhibitor, blebbistatin revealed an important role for the actinomyosin cytoskeleton in promoting clot retraction and thrombus stability. Furthermore, analysis of a range of signalling cascades involved in regulating platelet cytoskeletal function uncovered a major role for the Rho kinase signalling pathway in regulating thrombus contraction and stability.

Conclusions
These studies define a unique platelet contractile mechanism regulating thrombus stability under flow. This contractile mechanism is distinct from classical fibrin clot retraction in that the former is fibrin-independent, only occurs with larger thrombi and is regulated by shear. Moreover, unlike fibrin clot retraction, platelet thrombus contraction involves Rho kinase, a pathway not previously implicated in platelet contractile functions, raising the possibility that RhoA/Rho kinase inhibitors may represent new approaches to modulate thrombus stability in vivo.

Monday 15 October **1330-1500**
ANZSBT Symposium: New Technologies/Treatments **Central Room A**

1330

Granulocyte Transfusions

Jürgen Bux
German Red Cross Blood Service West & University of Bochum, Germany

Despite availability of antibiotics, antifungals and haematopoietic growth factors, infections remain a major threat to neutropenic patients. Although transfusion of granulocytes became more feasible with the development of cell separators, widespread use was hampered by low granulocyte yields, pulmonary transfusion reactions, and conflicting clinical study results. The introduction of the granulocyte colony-stimulating factor (G-CSF) in donor conditioning for granulocyte apheresis increased granulocyte yields. Transfusions of more than $2x10^{11}$ granulocytes are now routinely possible. In addition, G-CSF prolongs survival of transfused granulocytes by reducing their apoptotic rate, and improves granulocyte antimicrobial function. Gentle cell separation due to progress in apheresis technique and routine leucocyte cross-matching contributed to good tolerance towards granulocyte transfusions. However, required neutrophil dosage, frequency and right moment of granulocyte transfusion in a certain clinical condition have still to be determined in randomized multicentre studies.

Monday 15 October **1330-1500**
ANZSBT Symposium: New Technologies/Treatments **Central Room A**

1400

Electronic ID

Thomas Raife
University of Iowa Department of Pathology, Iowa City, IA, USA

Misidentification of patients, blood samples, and units of blood, remain among the most common and dangerous errors in transfusion medicine. Systems relying on manual labeling and documentation are inherently vulnerable to human error. Technology-assisted patient identification has become feasible but is not yet in wide use. With a grant from the U.S Agency for Healthcare Research and Quality, University of Iowa Hospitals replaced a manual Typenex patient identification system with a custom-built bar code label-based system in February 2005. Patients receive a bar coded wrist band from which bar code labels for blood typing specimens and requisitions are generated using a mobile wireless bedside computer. Blood bank bar code readers verify the match between the blood specimen bar code label and requisition bar code label. Blood units selected for the patient have bar code labels applied matching the specimen and requisition bar code. Prior to transfusion, the bedside computer is used to verify a match between the blood unit bar code label and the patient wrist band bar code. The system creates a closed loop of patient, specimen, and blood unit identification, and a complete audit trail for all steps, and has been enthusiastically adopted throughout the hospital. Incident reports have decreased by more than 80% and sample rejections have decreased by more than 90%. Errors and omissions detected by the system include mis-scans, skipped steps, wrong steps, and prevented identification errors of patients, samples, and blood products. Limitations of the bar code system have been identified, and the potential to further enhance the system using radio frequency identification devices (RFID) is under consideration.

Monday 15 October **1330-1500**
ANZSBT Symposium: New Technologies/Treatments **Central Room A**

1430

Blood Factories

Lars Keld Nielsen

Abstract not received at time of going to print

Monday 15 October **1330-1500**
Nurses Symposium: MDS **Meeting Room 5**

1330

Overview and Update on MDS

Julian Cooney

Abstract not received at time of going to print

Monday 15 October **1330-1500**
Nurses Symposium: MDS **Meeting Room 5**

1400

Logistics of Home Transfusion

Nicole Staples
Department of Haematology, Royal Perth Hospital, Perth, Western Australia

Aspects of haematology care are being delivered in the home setting as part of hospital-in-the-home programs. Examples include: chemotherapy, anticoagulation and IV antibiotics/antifungals. Home treatment of haemophilia patients with coagulation factor concentrates is well established. For some patients, subcutaneous immunoglobulin therapy is also administered at home. It is therefore timely to consider red cell and platelet transfusions in the home setting. Issues include:

- Severe transfusion reactions are uncommon, but can be managed more safely in hospital because of ready access to resuscitation equipment and staff. Evaluation of these risks, and their minimisation, must be addressed prior to establishing a home transfusion program.
- Approval from the hospital's transfusion committee, blood bank, and administration is required. Responsibilities of nursing, medical and blood bank staff need to be outlined, and policies and procedures in place for all aspects of the program.
- Nursing resources: requirement for suitably experienced nursing staff, with adequate time allocated per patient, in order to be present throughout entire duration of transfusion.
- Patient selection: eligibility and exclusion criteria need to be established. Examples of patients not suitable include those with: fever or active sepsis, cardiorespiratory compromise, red cell or platelet antibodies, previous transfusion reaction, or no previous transfusion.
- Site assessment of patient's home to ensure suitability e.g. distance from the hospital, cleanliness, telephone connection, family/caregiver support and ability to be present throughout transfusion.
- Patient consent: patient must have a clear understanding of risks of transfusions given at home compared to administration in hospital.
- Collection and labelling of pretransfusion specimens, including patient identification. Logistics of scheduling transfusion based on availability of FBP results and review; also component availability (platelets) and crossmatching (red cells).
- Issuing of blood component(s) by hospital blood bank, validated packaging and transportation.
- Administration of blood components. Issues to consider: venous access, patient identification and checking procedure, observations, documentation and disposal of blood bags.
- Management of complications. Requirement for clear plan of action if adverse event occurs. Access to medical advice from treating clinician, resuscitation equipment and protocols, including anaphylaxis. Patient access to 24 hour hospital liaison/back-up.

Monday 15 October **1530-1630**
HSANZ Symposium: New Approaches in the Lymphoproliferative Diseases
Arena 1B

1530

Immunomodulatory Drugs (IMiDs): Not Just for Myeloma Anymore

Myron C Czuczman

Abstract not received at time of going to print

Dr Myron Czuczman sponsored by Roche Products

Monday 15 October **1530-1630**
HSANZ Symposium: New Approaches in the Lymphoproliferative Diseases
Arena 1B

1600

Oncogenomics to Target Multiple Myeloma in the Bone Marrow Microenvironment

Kenneth C Anderson
Dana-Farber Cancer Institute, Boston, MA USA

Recent advances in genomics and proteomics in multiple myeloma (MM) increased understanding of disease pathogenesis, identified novel therapeutic targets, allowed for molecular classification, and provided the scientific rationale for combining targeted therapies to increase tumor cell cytotoxicity and abrogate drug resistance. Moreover, adhesion of MM cells to bone marrow stromal cells (BMSCs) upregulates genes for growth, survival, and drug resistance in tumor cells; adhesion molecules on MM and BMSCs; and cytokines in BMSCs. Thalidomide, lenalidomide, and Bortezomib are three agents which target the tumor cell in its microenvironment in both laboratory and animal models, and have rapidly translated from the bench to the bedside and FDA approval. Each achieved responses in relapsed refractory MM and then increased extent and frequency of response as well as prolonged progression free and overall survival, when used as initial therapy. Ongoing and future efforts are identifying next generation therapies in MM, and using oncogenomics to inform design of combination trials. Promising novel targeted therapies include agents targeting the tumor cell surface (CD40, CS-1, FGFR3), cytokines (VEGF, BAFF), and intracellular targets (MEK, PKC, NF-kB, Akt, IKK, cyclin D, proteasomes). Bortezomib has been combined with doxil (FDA approved), heat shock protein 90 inhibitors, Akt inhibitor perifosine, and lenalidomide; and lenalidomide with steroids, proteasome inhibitors, mTOR inhibitors, humanized monoclonal antibodies, and Akt inhibitors, based upon synergistic MM cell cytotoxicity in preclinical studies. Histone deacetylase inhibitor LBH589 and proteasome inhibitor Bortezomib block breakdown of ubiquitinated protein in the proteasome and aggresome, respectively, and trigger synergistic MM cell cytotoxicity *in vitro*, and combination clinical trials will begin soon. This new paradigm targeting the tumor cell in its microenvironment not only has great promise to change the natural history of MM, but also serves as a model for development of improved targeted therapeutics in cancer.

Dr Ken Anderson sponsored by Celgene Pty Ltd

Monday 15 October **1530-1630**
ANZSBT Symposium: Donors and Ethics **Central Room A**

1530

Ethical Issues in Collecting Blood from High Risk Populations: The South African and US Experience

Richard J. Benjamin
American Red Cross Blood Services, Washington, DC, USA

Donor Selection is a key safeguard for blood safety, allowing blood centers to drastically reduce the number of HIV- and hepatitis-infected blood units that are collected. Safety is achieved by deferring groups of donors who answer affirmatively on behavioral questions that are linked to increased risk. The majority of donors deferred due to high risk characteristics are not infected, and are therefore not dangerous. In general, society accepts that preventing these donors from donating is an acceptable practice in order to ensure blood safety for the majority of patients. Such discrimination becomes contentious when dealing with issues of race or sexual practice. This presentation will discuss the ethical issues around the use of race as a marker of risk in South Africa, where >11% of the population is infected with HIV-1 with a disproportionate racial distribution. This will be contrasted with the use of “male sex with males” (MSM) as a marker of risk in the U.S. An understanding of the ethical basis of donor deferral decisions may help drive better societal acceptance of policies that may appear to be discriminatory on initial examination.

Monday 15 October **1530-1630**
ANZSBT Symposium: Donors and Ethics **Central Room A**

1600

Psychology of Donation

Barbara Masser[1], Katherine White[2], Deborah Terry[1], Damon Cavalchini[3]
[1]University of Queensland, [2] Queensland University of Technology & [3]Australian Red Cross Blood Services, Brisbane, Queensland, Australia

There has been little theory-based research examining the psychosocial predictors of blood donation, especially repeat blood donation, within an Australian context. Using an extended Theory of Planned Behaviour (TPB; Ajzen, 1991) our program of research seeks to assess the role of attitudes, identity and norms in predicting blood donation within rural and urban settings and for repeat blood donation amongst early career donors. This research is being undertaken with a view to designing interventions to promote attitudinal and behavioural change for both blood donation initiation amongst current non-donors and repeat blood donation among early career donors. In our initial study undertaken with 820 donor and non-donor community members from Queensland, the results provided strong support for the extended TPB in the prediction of intention and actual behaviour. For both donors and non-donors, analyses implicated the key role that perceived control over the behaviour of blood donation plays for both intention to donate and actual donation behaviour. In addition, attitudes and anticipated regret (at not donating blood) were significant predictors of respondents' intentions to donate blood. A number of regional variations in predictors of intentions for donors and non-donors in terms of normative influences and identification were also noted. For established donors, structural equation modelling emphasised the important role that self identity as a blood donor plays in intention to donate blood, at the same time as giving some insights into how that key identity is formed. Preliminary findings from our second study which is tracking first time donors in Queensland over a two year period (Time 1 n = 1,958) will also be drawn upon to consider the different influences on the intentions and behaviour of non-donors, new donors and established donors.

Monday 15 October **1530-1630**
ASTH Symposium: Thrombosis Therapies **Central Room C**

1530

Venous Thromboembolism Prophylaxis

Harry Gibbs
Department of Vascular Medicine, Princess Alexandra Hospital, Brisbane, QLD Australia

Venous thromboembolism (VTE) is a common cause of morbidity and mortality. 50 – 70% of cases of VTE are caused by recent hospitalisation. Certain procedural and patient characteristics predict the development of VTE following hospitalisation and enable risk stratification. Effective methods to reduce VTE include the use of anticoagulants and mechanical leg compression. Anticoagulants that have been shown to be effective include unfractionated heparin, low molecular weight heparin, fondaparinux and warfarin; aspirin is relatively much less effective and should not be used as sole anticoagulant prophylaxis. Graduated compression stockings, intermittent pneumatic compression and foot impulse devices are effective mechanical forms of prophylaxis. Appropriate prophylaxis includes anticoagulant and mechanical prophylaxis for those at high risk and also the withholding of these agents where contraindications are present. The ANZ guidelines detailing risk stratification criteria and recommended prophylaxis will be presented. In spite of considerable data confirming the value of VTE prophylaxis for hospitalised patients, levels of appropriate prophylaxis are poor. Strategies for improving VTE prophylaxis include electronic or human alerts. Our approach has been to create the position of full time VTE prophylaxis clinical nurse consultant. The VTE nurse assists with VTE protocol creation and implementation and performs ongoing education, audit and feedback to maintain improvements. Appropriate VTE prophylaxis rates have improved from 28% to 77%. The sustainability of this approach is under ongoing assessment.

Monday 15 October **1530-1630**
ASTH Symposium: Thrombosis Therapies **Central Room C**

1600

Future Directions in Anticoagulation during Percutaneous Coronary Intervention

Darren Walters
The Prince Charles Hospital, Brisbane, QLD, Australia

An invasive strategy has become the preferred mode of treatment for patients with high-risk acute coronary syndrome. Adjunctive pharmacotherapeutic agents inhibiting platelet dependent thrombosis have improved the safety and efficacy of percutaneous coronary intervention (PCI) in these patients. Optimum anticoagulation however, remains a “Holy Grail” of interventional cardiology. A cocktail of medications that provide inhibition of both thrombin generation and platelet aggregation is required as no single agent has been developed to manage the thrombotic sequelae of coronary intervention especially in unstable coronary artery disease. Newer agents that provide improved and more selective inhibition of thrombin and platelet aggregation with a lower incidence of major bleeding are under clinical development. Direct thrombin inhibitors, novel P2Y12 receptor antagonists and direct Xa inhibitors may represent the future of anticoagulation in cardiac intervention.

Monday 15 October **1530-1630**
Nurses Symposium: the "Fall Guy" in Haem/BMT **Meeting Room 5**

1530

Tissue Typing and Sibling Donors

Mary McGurgan[1], Cassandra Reid[2]
[1]*Blood and Marrow Transplant Service, Westmead Hospital, Sydney, NSW, Australia*
[2]*Haematology Department, Royal North Shore Hospital, Sydney, NSW, Australia*

Finding a suitable donor is a challenge in the specialty of Allogeneic Haemopoietic Stem Cell Transplantation (HSCT). 1 in 3 patients who would benefit from HSCT will have a suitably matched sibling donor. There are clearly defined guidelines for the recruitment and management of Volunteer Unrelated Donors. Is this the case for sibling donors?

BMT Co-ordinators from two Allogeneic HSCT centres will present their centres' current practice on the recruitment and management of sibling donors. Case scenarios highlighting the myriad of sibling donor issues will be presented.

This will be an interactive session and the audience will be invited to share their experiences.

Monday 15 October **1530-1630**
Nurses Symposium: the "Fall Guy" in Haem/BMT **Meeting Room 5**

1600

Remote Area Issues – A Haematology Nursing Perspective

Kris Liebke
Cancer Care and Haematology Unit, Lismore Base Hospital, Lismore, NSW, Australia

The prospect of a bone marrow or stem cell transplant is daunting for anyone, but for regional patients there are many added issues. Patients are relocated many hours from their homes, away from family, friends and other support networks. Other issues such as finances, transport, employment and navigating through the health system are also often exacerbated for the regional patient.

There are also many challenges for regional / rural nurses responsible for coordinating transplant patients. Establishing and maintaining good two-way communication channels with transplant centres, discovering the 'tricks of the trade', arranging transport and accommodation for patients who have limited travel / city experience and coordinating treatment schedules to allow for things such as quiet times for small business or a macadamia nut harvest.

This presentation will offer insight into some of these issues. It will present case samples of real people and the challenges they have experienced as regional patients entering metropolitan services. It will review how we work well together in our regional / metropolitan health care team relationships and where things could be improved.

Monday 15 October **1730-1830**
Masterclasses **Meeting Room 5**

HSANZ
Controversies in Multiple Myeloma

Jean-Luc Harousseau
Department of Hematology, University Hospital Hotel-Dieu, Nantes, France

Abstract not received at time of going to print

Dr Jean-Luc Harousseau sponsored by Pharmion

Monday 15 October **1730-1830**
Masterclasses **Meeting Room 6**

HSANZ
Modern Management of GVHD

Robert Negrin

In this Master Class we will attempt to take the lessons learned from animal models and apply them to the prevention and treatment of Graft vs Host Disease. We will examine the current treatment strategies and discuss how they could be improved and assessed.

Monday 15 October **1730-1830**
Masterclasses **Meeting Room 4**

HSANZ
The Evolution of Monoclonal Antibody Therapy for B-cell Lymphoma

Myron C Czuczman

Abstract not received at time of going to print

Dr Myron Czuczman sponsored by Roche Products

Monday 15 October **1730-1830**
Masterclasses **Meeting Room 8**

ANZSBT
TRALI

Christopher C Silliman
Bonfils Blood Center and Dept. of Pediatrics, University of Colorado School of Medicine, Denver, CO, USA

The Masterclass will consist of an informal discussion with regard to 1) a summary of the generation of biologic response modifiers in blood components and their role in TRALI and 2) the pathophysiology of TRALI and the similarity about ALI from both antibodies and other biologic response modifiers.

Monday 15 October **1730-1830**
Masterclasses **Meeting Room 9**

ANZSBT
Molecular Studies of Blood Groups

Geoff Daniels
Bristol Institute for Transfusion Sciences, NHSBT, Bristol, UK

The molecular genetics era of blood groups began in 1986 with the cloning of the gene for the MN protein. Now the genes for all but one of the 29 blood group systems have been cloned and the molecular backgrounds to almost all clinically important blood group polymorphisms have been identified. This makes it possible to predict blood group phenotypes from genomic DNA with a high level of accuracy. Most of the blood group polymorphisms arise from single nucleotide polymorphisms (SNPs), though other mutations or SNPs elsewhere in the gene or in another gene can affect antigen expression. The Rh system is very complex, with a large variety of RhD variants resulting from different mutations in *RHD* and *RHCE*.

The main applications of molecular genetics to red cell blood grouping include: the prediction of fetal blood groups in pregnant women with blood group antibodies in order to determine whether the fetus is at risk from HDN; prediction of clinically important blood groups of multiply-transfused, transfusion-dependent patients so that matched blood can be provided to assist in the prevention of blood group antibody production; defining RhD and RhCcEe variants, predicting blood group phenotypes on red cells coated with immunoglobulin (DAT+); and confirming zygosity in the production of reagent red cells.

High-throughput methods for detecting the SNPs and other gene sequence variations associated with blood group polymorphism will make it possible to genotype most donors and patients for all clinically significant blood groups, enabling electronic matching, which would reduce immunisation and the prevalence of haemolytic transfusion reactions. The Bloodgen project in the UK has led to the development of a DNA microarray for 176 allelic variants from 10 blood group systems, including 87 *RHD* genotypes, which has a very high level of accuracy for predicting blood group phenotypes.

Monday 15 October **1730-1830**
Masterclasses **Meeting Room**

ASTH
Diagnosis and Management of Platelet Disorders

Alan T Nurden and Paquita Nurden
Centre de Référence des Pathologies Plaquettaires, Plateforme Technologique et d'Innovation Biomédicale, Hôpital Xavier Arnozan, Pessac, France

Defects in platelet production or platelet function compromise the ability of platelets to assure hemostasis and can result in bleeding. It is not always easy to make a correct diagnosis. We still receive patients with inherited thrombocytopenias who have been incorrectly diagnosed with immune thrombocytopenic purpura and have received inappropriate treatment. An essential international task is to conceive standardized diagnostic algorithms. Consultation should establish the family history and the nature, frequency and severity of bleeding. Associated clinical conditions such as deafness, renal disease may indicate a specific genetic disorder and should be noted. Platelet counts are essential to diagnosis; when low, smears should be examined for (i) giant platelets missed by electronic counters and (ii) granule content. Electron microscopy can provide specialist information on platelet morphology. Inherited thrombocytopenias may include VWD-type2B and platelet-type VWD whose frequency may be underestimated. Platelet function testing mostly concerns the turbidometric evaluation of platelet aggregation in citrated platelet-rich plasma with a series of agonists including ristocetin, ADP, collagen, epinephrine, arachidonic acid and thrombin-receptor activating peptide (TRAP). Follow-up examinations are required to confirm initial results and to permit selected testing of defective pathways. Simultaneous measurement of ATP secretion and aggregation will highlight storage pool disease. Membrane receptors such as GPIb-IX-V and αIIbβ3 are evaluated by flow cytometry, as is the capacity of agonists to (i) activate αIIbβ3 and (ii) generate procoagulant activity. Devices such as the PFA-100 confirm the presence of defective platelet plug formation but progress is needed in evaluating thrombus formation under flow. Treatment prior to surgery can involve increasing VWF levels with desmopressin, but the main response to bleeding is to transfuse platelet concentrates although the presence of inhibitors (antibodies) needs surveillance. Promoting fibrin formation with recombinant FVIIa is an increasingly used alternative. A somatostatin analog (octeotride) is proving useful to treat angiodysplasia.

Monday 15 October **1730-1830**
Masterclasses **Meeting Room 3**

Nursing Programme
Haematology and Palliative Care

Pam McGrath
International Program of Psycho-Social Health Research (IPP-SHR), Central Queensland University, PO Box 1307, Kenmore Qld, Australia

Purpose
Research indicates that end-of-life care in haematology is most commonly situated in the curative system and is associated with the distress of escalating technology without appropriate referral to palliative care. A recent two-year study, funded by the National Health and Medical Research Council (NHMRC), addressed these problems through the development of a trilogy of models of care (Functional, Evolving and Refractory) detailing the factors that facilitate and obstruct the integration of haematology and palliative care. The masterclass explores findings from the study that highlight the important role of nurses in providing leadership in this sensitive and important area of patient and family care.

Methods
The qualitative methodology used open-ended interviews (n=90) that were audio-taped, transcribed verbatim and thematically analysed. The three-stage process for model development is based on the National Cancer Control Initiative's methodology for Optimising Cancer Care in Australia. The trilogy of models has been subject to rigorous peer review by a national panel of haematology and palliative care experts.

Results
The findings indicate that there are three distinct models in end-of-life care in haematology in Australia: (a) the Functional model; (b) the Evolving Model; and (c) the Refractory Model. The masterclass details the characteristics of each model of service delivery and elaborates on the role of nurses as key agents of positive change towards the appropriate integration of palliative care in haematology.

Conclusion
The outcome is the initiation of a dialogue about the role of leadership that nurses can and do provide in ensuring that haematology patients receive best practice end-of-life care. Copies of the booklet for health professionals developed from the study, titled 'Haematology and Palliative Care: Towards an Integrated Practice', will be made available at the presentation for interested delegates.

Notes: