Rejuvenation of Allogeneic Blood

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No Disclosures

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 Cardiac surgery has the highest allogeneic blood use

 Allogeneic blood at tertiary centers is often close to expiry (42 days)

Older blood = Increased Morbidity/Mortality



Li et al 2008

The Storage lesion

 "Myriad of biochemical and morphological lesions that occur during storage" (D'Alessandro et al 2017)



Effects of Storage on the RBC

• Results in shape change (echinocytosis)





- Membrane loss
- Loss of about 20% of cell surface and an increase in cell density
- Increased osmotic fragility
- Rheologic (flow dynamic) changes
- Results in increased red blood cell fragility and impaired function
- Increased hemolysis
- Increased aggregation
- Significant reduction in deformability (Zubair 2009)

Capillary Flow Dynamics



Compared with fresh blood, transfusion of Stored RBC's results in:

- •Decreased flow velocities
- Increased aggregation
- •Endothelial adherance
- •Increased leukocyte rolling
- •Regional tissue ischemia
- •Statistical significance P<0.05 Arslan *et al* (2005)

Transfusion of older RBC's results in extra and intravascular hemolysis



Hod et al 2011

IMMUNOMODULATORY EFFECTS

- Exogenous antigenic proteins, have numerous pro and anti-inflammatory effects
- Human Leukocyte antigen (HLA) proteins
 Improved graft survival
 - Reduction in inflammation in Crohn's disease.
- Fas Ligand

Activation/neutralisation of Natural Killer cells

(Barshtein et al 2011)

The Iron hypothesis



Hemolysis and Nitric Oxide

 "Adminsitration of stored allogeneic blood results in hemolysis, represented by significant increases in plasma free hemoglobin"

• "During intravascular hemolysis diffusional barriers are disrupted, resulting in efficient nitric oxide scavenging and endothelial dysfunction"

Rother et al 2005

Significance of Plasma free Hemoglobin

Nitric Oxide synthesis

NO Signaling

Proinflammatory effects

Oxidative stress

Platelet aggregation

Smooth muscle Dystonias

Impaired Autoregulation of blood flow



Russel et al 2005

Storage Medium for RBC's

- Not Ideal!!
 - Acidic (ph 5.6-5.8)
 - Energy resources quickly deplete
 - Buffering capacity of the RBCs is soon exhausted
 - Enzymatic function is altered
 - Lactic acid increases over time
- Blood services "could benefit the advancement of RBC storage systems"
- Huge financial burden for companies to "obtain licensure and to bring a new RBC storage system to a market"

Sparrow 2012

Rejuvesol

- Sterile non pyrogenic solution:
 - sodium pyruvate
 - inosine
 - adenine



- mono- and dibasic sodium phosphate

• *In vitro* processing solution for the rejuvenation of red blood cells (RBCs)

Rejuvesol

- FDA approved
- Restores:
 - 2,3-diphosphoglycerate (2,3-DPG)
 - Adenosine triphosphate (ATP)
- Reversal of Echinocytosis (cell membrane integrity)

Restoration of RBC deformability

 Recommended for Allogeneic blood after 14 days storage





RBC Rejuvenation is not a New Concept

Improved Oxygen Delivery to the Myocardium during Hypothermia by Perfusion with 2,3 DPG-Enriched Red Blood Cells

1970

C. R. Valeri, M.D., M. Yarnoz, M.D., J. J. Vecchione, M.D., R. C. Dennis, M.D., J. Anastasi, D. A. Valeri, L. E. Pivacek, H. B. Hechtman, M.D., C. P. Emerson, M.D., and R. L. Berger, M.D.

TREATMENT OF SEVERE HYPOXIA WITH RED CELLS HIGH IN 2, 3-DIPHOSPHOGLYCERATE

H. J. PROCTOR, M.D., F.A.C.S., JOHN C. PARKER, M.D., F.A.C.S., JAMES FRY, B.A., AND GEORGE JOHNSON, JR., M.D., F.A.C.S.

From the Department of Surgery, University of North Carolina School of Medicine, Chapel Hill 1973

Transfusion of 2,3 DPG–Enriched Red Blood Cells to Improve Cardiac Function

Richard C. Dennis, M.D., Herbert B. Hechtman, M.D., Robert L. Berger, M.D., Louis Vito, M.D., Richard D. Weisel, M.D., and C. Robert Valeri, CAPT, MC, USNR 1977

Rejuvesol, CPB and ECMO

BLOOD COMPONENTS

Rejuvenation improves roller pump-induced physical stress resistance of fresh and stored red blood cells

Monique P. Gelderman and Jaroslav G. Vostal

2011

Hemolysis is reduced in



2-3 DPG restoration

Oxygen delivery

Old RBC's= High affinity for O₂
Reduced ability to dissociate

Clinical effects= SVO₂
Oxygen delivery at capillaries

Raat et al 2009

Raat et al 2009

• "Stored human RBC's (5-6 weeks old) show a deficit in the oxygenation of the microcirculation"

 "Look at the effect of rejuvenation of stored RBCs on their ability to deliver oxygen in a rat kidney"

Improved Oxygen delivery

Raat et al 2009

Oxygen delivery

• "Lower oxygen affinity-high 2,3 DPG group had improved oxygen delivery to tissue"

 "Prolonged storage results in lowering capillary perfusion and impaired oxygen delivery"

Potential clinical applications

- Rejuvenation of RBC's for anemic patients prior to Cardiac surgery
- Bed side rejuvenation for Post Cardiac surgery/ECMO patients
- Reduction in blood transfusions associated with hemolysis induced anemia
- Better oxygen delivery/consumption

ECMO and Hemolysis

Cavitation

Multiple blood products

Duration of ECMO

High centrifugal negative inlet pressures

Venous chatter

Activation of clotting cascade

High sheer stress

Foreign surface

- Hemolysis

Kumar et al 2010

ECMO and PfHb

 "Plasma Free Hemoglobin Is an Independent Predictor of Mortality among Patients on Extracorporeal Membrane Oxygenation Support"

Omar 2015

Clinical case study

Norepinepherine day 1: 4mcg/ml Norepinepherine day 12: 40mcg/ml 0.04 mcg/ml Vasopressin Methylene blue given

Organ rejection

 "In heart failure patients undergoing implantation of ECMO or ventricular assist devices, multiple transfusions are common and their immunogenicity is directly related to the number of units given "

Scornika and Meier-Kriesche 2011

Would Rejuvenation helped??

- Further investigation needed
 - Massive transfusion
 - Hemolysis?
 - Allogenic blood/ ECMO circuit??
- Better oxygen delivery?
- More robust RBC's?
 Reduction in transfusion requirments?
- Prevention of infection?

Monitoring ECMO patients

• Difficult to independently attribute morbidity and mortality to high hemolysis

• Omar et al (2015) "propose the routine checking of PfHb 24-hours after ECMO initiation for early identification and treatment of the cause of hemolysis"

Potential Benefits of rejuvenation for ECMO patients

- Oxygen delivery 12-3 DPG
- Reversal of Echinocytosis
- Reduced inflammatory response
- Reduced infection rates?
- Reduction in RBC transfusion rates?
- Heart transplant candidacy
- Lower Plasma Free Hemoglobin
- Reduction in hospital length of stay???

Questions/Comments