Transfusion of red blood cells is common in patients undergoing cardiac surgery. The physiological and clinical consequences of anemia are well documented, with several studies demonstrating an association between decreasing hemoglobin concentration and adverse outcome. As the degree of anemia increases, the normal physiologic response is to increase organ flow (cardiac output) and/or increase oxygen extraction. At some point, these compensatory mechanisms become exhausted and a state of inadequate oxygen delivery for tissue needs is reached. Although some studies have suggested that tissue hypoxia does not occur in the brain and heart until the hemoglobin (Hb) is below 4 g/dL, clear evidence of tissue hypoxia occurs earlier in other tissues. In addition, activation of hypoxic mechanisms and increased incidence of morbidity and mortality are observed near a hemoglobin of 6-7 g/dL. There are limited options for treating anemia, and transfusion of stored red blood cells (RBCs) has been the cornerstone of management of patients with life-threatening anemia. RBC transfusions are usually administered to improve tissue oxygen delivery. Whether or not they do so depends on a variety of factors, including the conditions and length of storage, and the clinical circumstances in which they are administered. Transfusion decisions should always be based on an evaluation of whether the risks of transfusion outweigh the risks of anemia. Balance of risk may be influenced by physiologic, pathophysiologic and pharmacologic factors. For example, patients with neurotrauma and acute coronary syndromes may benefit from a higher Hb than other patients. Also, recent data have suggested that bleeding increased the risk of stroke in beta blocked patients. Treatment is dependent on the individual patient and clinical parameters.

**Measurement of oxygen delivery:**
A variety of physiologic measurements and monitoring techniques have been used to measure tissue oxygen delivery and the ‘critical’ point beyond which harm can occur or benefit of transfusion can accrue. These include calculation of oxygen content, flow, and consumption, measurement of hemodynamic variables, venous oxygen saturation/content, and direct monitoring of tissue oxygenation using implanted oxygen electrodes, transcutaneous probes, phorsphorescence quenching, or near infra-red spectroscopy.

**Effect of Storage on RBCs:**
The shelf life of RBCs is determined exclusively by a minimum 24-hour post-transfusion survival of 75% of the transfused red cells. Additional criteria are related to free hemoglobin (<1.0%) and white blood cell levels (<1 million/unit in leuko-reduced blood). Improvements in storage solutions and techniques have increased the shelf-life of blood from 21 to the current 42 days. However, this means that a patient who receives 4 units of packed RBCs may be receiving 1 whole unit of dead cells. It has been estimated that 20-40% of all stored RBC units are >28 days old at the time of transfusion, and with supply fluctuations, up to 40% of blood may be within 3 days of expiration.

RBCs undergo many complex biochemical, environmental, structural and biomechanical changes during storage. Because of the storage medium, the RBCs are subjected to a lower pH and higher glucose concentration within minutes of storage, and these continue to change slowly for the duration of storage. Within hours to days of storage, there are progressive changes in 2,3
DPG, potassium, lactate, free hemoglobin, RBC ATP, pO\textsubscript{2}, Hb O\textsubscript{2} saturation, antioxidant status and RBC deformability. Within 2-3 weeks the 2,3 DPG levels approach zero. By the end of 6 weeks, although the pO\textsubscript{2} is well over 100 mmHg and the Hb O\textsubscript{2} saturation near 100%, the potassium is over 20 mmol/L, and the lactate is greater than 15 mmol/L. The shape of the red cell is transformed from a normal biconcave disc to a spiculated and crenated echinocyte, then to a swollen spherococyte. No significant changes occur in other electrolytes (chloride, calcium, magnesium), RBC phosphatidyl serine expression, or RBC adhesion to endothelial cells. In a recent elegant series of experiments, Bennett-Guerrero et al demonstrated a rapid and marked fall in 2 bioactive forms of RBC nitric oxide (Hb-bound NO, and S-nitrosohemoglobin), as well as their effector response, RBC mediated hypoxic vasodilation.

### Storage Effects on Packed Red Cells

<table>
<thead>
<tr>
<th>Increase with Storage</th>
<th>No Change with Storage</th>
<th>Decrease with Storage</th>
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</thead>
<tbody>
<tr>
<td>pO\textsubscript{2}</td>
<td>Chloride</td>
<td>ATP</td>
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<tr>
<td>Free Hb</td>
<td>Calcium</td>
<td>pH</td>
</tr>
<tr>
<td>Potassium</td>
<td>Magnesium</td>
<td>2,3-DPG (1\mu mol/gHb – @ 21d)</td>
</tr>
<tr>
<td>Lactate</td>
<td>Met Hb</td>
<td>Hb-Bound NO</td>
</tr>
<tr>
<td>Hb Saturation</td>
<td>RBC Surface PS Expression</td>
<td>SNO-Hb</td>
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<tr>
<td>Cytokines (variable)</td>
<td>RBC Surface to volume ratio</td>
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<tr>
<td>Glucose</td>
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<td>MCF</td>
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<td>MCV</td>
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<td>Sodium</td>
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<td>RBC Osmotic Fragility</td>
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<tr>
<td>RBC Deformability</td>
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</tbody>
</table>

**Consequences of Transfusion of stored RBCs: Laboratory Studies**

The mechanisms by which prolonged RBC storage produce adverse clinical outcomes are likely related to impaired oxygen delivery or upregulation of the inflammatory response. 2,3 DPG affects the oxyhemoglobin dissociation curve, and although it is regenerated after transfusion, complete restoration can take 2-3 days. The cause of 2,3 DPG depletion is thought to be both because of the low pH of the storage solution but also as a result of glucose metabolism before refrigeration. Several studies have demonstrated reduced microvascular tissue oxygenation with stored blood. For example, Tsai et al have shown that replacing 25% of circulating RBCs with 28 day old cells reduced microvascular flow and functional capillary density by over 50%. Tissue pO\textsubscript{2} with the old blood was 3.5 mmHg compared to 14.4 mmHg with fresh RBCs. Critical oxygen delivery is lower in animals who received transfusion of fresh whole blood. Similarly, Rigamonti et al found that fresh blood produced a greater increase in brain tissue pO\textsubscript{2} and cerebral blood flow than stored blood in a rat model of acute fluid resuscitation.

**RBC Transfusion alternatives:**

Several artificial oxygen carriers have been developed as ‘blood substitutes’. Many studies with either perflourocarbon or hemoglobin based oxygen carriers have shown these products to be as effective or better than blood at improving oxygen delivery or tolerating to critical oxygen delivery. However, clinical use of these products has been restricted by either safety concerns or insufficient clinical development to date.

**Consequences of Transfusion of stored RBCs: Clinical Studies**

Numerous reports have identified an association of RBC transfusion with adverse clinical outcomes. The majority are retrospective or prospective observational studies which use varying degrees of multivariable analysis to reduce the possible confounder that sicker patients are the
ones who receive transfusion. There are a few randomized trials and no specific systematic reviews as yet. Prolonged storage time has been associated with increased morbidity and mortality in a wide variety of clinical settings, including patients with trauma, sepsis, and critical illness. For cardiac surgery, length of RBC storage has been correlated with death, renal dysfunction and ICU length of stay. It has been estimated that each additional day of RBC storage increases the risk of pneumonia by 1-6%. However, Weiskopf et al found no difference in the ability of fresh or stored blood to restore brain oxygen deficits induced by anemia in humans, and at least 2 observational studies in cardiac surgical patients have not found an association between duration of storage and increased morbidity. Nonetheless, Leal-Noval et al found that transfusion of blood stored less than 20 days significantly increased brain tissue oxygenation in patients with severe traumatic brain injury, whereas blood stored 20 days or longer did not.

There have been few randomized clinical trials done examining the age of blood on physiologic or outcome parameters. Two small studies (n=22-23) monitored gastric pH – one found an inverse relationship between age of RBCs and gastric pH, whereas the other found no effect on pH or global indices of tissue oxygenation. The differences may be due to the patient population studied or the use of non-leukoreduced blood in the study with a positive effect. A small pilot RCT of RBC storage in 57 patients, the majority of whom had cardiac surgery, found higher but not significant differences in the fresh blood group in terms of death or life-threatening complications. Similarly, neonatal trials have been relatively small. One RCT reported that the use of fresh whole blood in the pump prime of 200 infants undergoing CPB offered no advantage compared to packed red cells+plasma. In fact, the patients in the fresh whole blood group had a longer length of stay and higher fluid balance. There currently is a randomized trial underway in Canada. The ARIPI (Age of Red Blood Cells in Premature Infants Study) is enrolling 450 premature infants with a birthweight <1250 gm to study a primary outcome of mortality and/or composite outcome of necrotizing enterocolitis, intraventricular hemorrhage, bronchopulmonary dysplasia and retinopathy of prematurity.

The largest study examining the effect of RBC storage was recently published by Koch et al in the New England Journal of Medicine. They studied over 6000 patients undergoing cardiac surgery at the Cleveland Clinic who received either new (<14 day storage) vs old (>14 day) RBC transfusions. Using regression analysis and propensity scoring to balance some pre-existing population imbalances, they found that patients given older blood had significantly worse composite clinical outcomes consisting of renal failure (2.7% vs 1.6%), prolonged intubation (9.7% vs 5.6%), and sepsis 4.0% vs 2.8%). In addition, in-hospital and 1-year mortality were significantly higher in the old-blood group.

**Conclusions and Future Implications:**
Although improvement of tissue oxygen delivery is usually a desired goal of red cell transfusion, this goal is often not demonstrable, either because of the physiologic conditions of the patient, the characteristics of the blood or the measurement techniques used. There is ongoing debate about the best Hb trigger for transfusion. In addition, there is increasing evidence that duration of storage of blood reduces the quality of transfused RBCs and may adversely affect physiologic parameters and clinical outcome. However, limiting transfusions to only fresh (ie <14 days) RBCs would have significant implications for an already strained blood system. Blood conservation strategies should continue to be a high priority for patients at risk for transfusion. Future efforts to improve the viability of red cells in innovative storage solutions and conditions may yield RBCs with survival rates closer to 90%, even with prolonged storage. Strategies to
restore RBC NO and S-nitrosohemoglobin could improve physiologic and rheologic properties of RBCs and increase their survival. Better understanding of the measurement of and mechanisms by which transfusion affects tissue oxygen delivery should help optimize management of patients at risk of anemia and transfusion.

Reference List


