Blood transfusion has been a mainstay of medical therapy for vast numbers of patients worldwide for several decades. Nearly 15 million units of packed red blood cells (RBCs) and whole blood are transfused annually in the United States alone. Until recently, the major risks from blood transfusion were thought to be transmission of viral infections, and overall blood transfusion was believed by most providers to be safe. Corwin and colleagues demonstrated in 1995 that blood transfusion in the intensive care unit (ICU) was common, and often without clear indication.

In 1999 Hebert and colleagues published the landmark TRICC (Transfusion Requirements In Critical Care) trial, a multicenter, randomized controlled clinical trial that demonstrated that a restrictive transfusion strategy was not only safe, but in certain patients possibly beneficial. Much work has been done since that time to further the understanding of the pathophysiological effects of red cell transfusion, but there is still a lack of clear guidelines for transfusion of packed red blood cells (PRBCs) in settings other than the actively hemorrhaging patient.

Current guidelines recommend a transfusion threshold hemoglobin level between 7 and 8 g/dL in noncritically ill patients. A notable exception are recommendations for transfusion during early sepsis, based largely on the study by Rivers and colleagues demonstrating improved outcomes in septic patients with early and aggressive measures to improve oxygen delivery.

Animal and human studies suggest that although certain patients may tolerate very low hemoglobin levels, anemia in others may be harmful. However, it is not clear at what level of hemoglobin, and in what patients, transfusion is beneficial.

The notion that transfusing RBCs will acutely increase tissue oxygenation in sepsis is predicated on several assumptions: first, that tissue and cellular oxygenation are inadequate; second, that RBC transfusion improves tissue oxygen availability; third, that cells will be able to use the delivered oxygen; and lastly, that the benefit of transfusing the red cells outweighs the risk to the individual patient.
Increasing hemoglobin concentration by transfusion increases global oxygen delivery as measured by systemic variables, but little is known about whether transfusion provides an increase in the oxygen delivery to tissue at the microvascular level.8

PRINCIPLES OF OXYGEN TRANSPORT

All cells require oxygen for aerobic metabolism to maintain normal cellular function. Because oxygen cannot be stored in the cells, a constant supply that matches the metabolic needs of each cell is required.9 Oxygen itself is poorly soluble in water and plasma, and is transported to tissues via hemoglobin.

There are four critical steps in the chain of oxygen transport: (1) delivery of oxygen to the lung; (2) diffusion into the blood, where it binds rapidly and reversibly to hemoglobin within the RBC; (3) transportation to the various tissues of the body by blood flow; and (4) diffusion down a concentration gradient into the tissue, and ultimately to the mitochondria of each cell.9–11

Oxygen demand differs markedly between and within specific organs.12 Oxygen delivery is matched to local need by adjustments of vascular tone in the macro- and microcirculation,13 which are under the control of autonomic signals and local metabolic stimuli.11–13 These regulatory mechanisms are still not well understood, and even in the presence of regional cellular hypoxia, an increase in systemic oxygen delivery may not result in greater oxygen availability to the cell.13 Once delivered, oxygen diffusion into the tissue is determined by the oxygen tension gradient between the blood and the tissue.14,15

Systemic oxygen delivery (DO2) is dependent on cardiac output (CO) and arterial oxygen content (CaO2)10,13,14:

\[
DO_2 = CO \times CaO_2
\]

Arterial oxygen content is the sum of hemoglobin-bound and dissolved oxygen.10

\[
CaO_2 = (Hb)(1.34)(SaO_2) + (0.0031)(PaO_2)
\]

where 1.34 is the amount of oxygen carried by 1 g of hemoglobin, Hb is the hemoglobin concentration (grams per 100 mL), SaO2 is the arterial oxygen saturation, 0.0031 is the solubility of oxygen in the plasma, and PaO2 is the partial pressure of oxygen in the blood.

Under normal physiologic conditions, dissolved oxygen in plasma and cells contributes minimally to the total oxygen content of the blood.10,12 In nonanemic patients with a hemoglobin level of 14 g/dL and breathing room air, less than 2% of the arterial oxygen content is dissolved in the plasma. By contrast, in a severely anemic patient with a hemoglobin level of 5 g/dL and breathing 100% oxygen, 20% of the total arterial oxygen is dissolved in the plasma.16,17

At the cellular level tissue oxygenation is in part determined by the oxyhemoglobin dissociation curve.12,18 The characteristic sigmoid shape of the oxyhemoglobin dissociation curve results from a complex interaction between hemoglobin subunits reflecting a nonlinear affinity for oxygen binding.14 This binding affinity may be altered by pH, temperature, and RBC 2,3-diphosphoglycerate (2,3-DPG) levels.17

In health, the amount of oxygen delivered to the whole body exceeds resting oxygen requirements by a factor of two- to fourfold.17 In a normal resting adult at sea level, \( DO_2 \) is approximately 1000 mL/min based on a cardiac output of 5 L/min, a hemoglobin level of 15 g/100 mL, and \( SaO_2 \) of 100%.9 Under these conditions only about 25% (250 mL/min) of the oxygenated hemoglobin will be deoxygenated, providing a large
physiologic reserve. This ratio of oxygen consumption (VO₂) to DO₂ is termed the oxygen extraction ratio.

As hemoglobin levels fall, oxygen consumption remains unchanged until a critical DO₂ (DO₂(crit)) is reached, where cardiac output and oxygen extraction compensation can increase no further, and oxygen consumption (VO₂) begins to drop. In other words, VO₂ is limited by demand above critical DO₂ and by supply below it.

In animal studies of euvoletic hemodilution, it has been shown that the hemoglobin level corresponding to DO₂(crit) is approximately 3 to 3.5 g/dL. This level is similar in healthy humans. Of note, the microcirculation, where oxygen is delivered, is carefully regulated to a hematocrit of 12% to 15%, which corresponds to the above DO₂(crit).

CAUSES OF TISSUE HYPOXIA

Tissue hypoxia can be caused by decreased DO₂ from decreased hemoglobin concentrations (anemic hypoxia), decreased cardiac output (stagnant hypoxia), or low hemoglobin saturation (anoxic hypoxia). A fourth term, cytopathic hypoxia, was coined by Fink. Cytopathic hypoxia refers to a state of cellular hypoxia whereby generation of adenosine triphosphate (ATP) by means of aerobic metabolism is limited not by the availability of oxygen, but by alterations in the cellular elements needed to perform oxidative phosphorylation; this is believed to be the prevalent form of hypoxia in sepsis. Anoxic hypoxia can also be related to abnormalities in oxyhemoglobin dissociation and the ability of the RBC to traverse the microcirculation.

MEASUREMENT OF TISSUE OXYGENATION

Shock is the point at which DO₂(crit) is reached and cellular respiration is compromised. Clinical evaluation is frequently used at the bedside to assess shock. Commonly used markers include pulse rate, blood pressure, skin temperature, and urine output. These markers are unreliable, can be slow to change in the setting of acute shock, and may become apparent only in later stages of shock. Laboratory markers such as base deficit, serum lactate, anion gap, and pH are also used to monitor impaired tissue oxygenation. Although these measurements have been shown to be predictive of poor outcome, increased mortality, and multiple organ failure, they are global measures that may obscure the presence of significant changes in oxygen uptake in individual organs.

In sepsis there is marked variation in tissue oxygenation, and systemic oxygenation parameters often do not reflect local tissue oxygen levels. Two recent studies in septic patients demonstrated that global parameters of hemodynamics and oxygenation had no correlation with measured changes in microvascular perfusion or tissue saturation.

Determination of adequacy of tissue oxygenation ideally requires local organ measurements, but at present there are few clinical techniques to directly monitor tissue oxygen levels or quantify tissue oxygen consumption. Methods such as gastric tonometry and near-infrared spectroscopy (NIRS) have been developed, and although promising, none has yet been validated for widespread clinical use.

Measured CaO₂ and cardiac output are commonly used to assess global DO₂. Errors in measurements are common, especially in critically ill patients. The accepted error of cardiac output measured by thermodilution is approximately 10%. However, this error tends to be larger at extremely high or low values, such as those commonly observed in patients with septic or hypovolemic shock.
respectively. When pulmonary artery catheter-derived measured are used according to the inverse Fick method, both oxygen delivery and oxygen consumption use measurements of cardiac output and arterial oxygen content in their calculation. By a phenomenon known as mathematical coupling, this calculation may create the impression that cardiac output and oxygen consumption are positively correlated across a wide range of values. Previous studies have suggested that in critically ill patients, interventions that increase DO₂ also appear to increase VO₂, creating a more linear relationship between DO₂ and VO₂ than that seen in healthy subjects. This “oxygen supply dependency,” however, is highly artificial and confounded by mathematical coupling. In studies in which VO₂ has been measured independently by indirect calorimetry, the apparent supply dependency is not seen.

Mixed venous (or central venous) oxygen saturations are used clinically to monitor systemic DO₂, with the assumption that low levels imply inadequate oxygen supply. Central venous oxygen saturation is a recommended marker for evaluating adequacy of resuscitation in early sepsis. Oxygen content in a mixed or central venous blood sample is a blood flow–weighted average of venous oxygen content of different organs, making it an insensitive marker of tissue hypoxia in individual organs. Blood transfusion increases mixed venous oxygen saturation, but may have no effect on tissue oxygenation. Stored erythrocytes tightly bind oxygen so mixed venous oxygen levels will be higher, even though tissue oxygenation may be unchanged. Therefore, despite current recommendations, mixed venous oxygen saturation is actually a poor indicator of tissue anoxia.

Blood lactate levels are also commonly used clinically for monitoring systemic DO₂. An elevated lactate implies inadequate tissue oxygenation and conversion to anaerobic metabolism. However, regional tissue hypoxia can be significant in the presence of normal lactate levels.

**PHYSIOLOGIC RESPONSE TO ACUTE ANEMIA**

As long as euvolemia is maintained, normal physiologic compensatory mechanisms allow most critically ill patients to tolerate anemia surprisingly well. There are few data regarding how the normal physiologic adaptive responses are affected by various disease states, but presumably compensatory mechanisms are less effective in people who are ill or elderly.

With acute normovolemic anemia, the reduction in arterial oxygen content is compensated for by (1) increasing the cardiac output, (2) altering the distribution of blood flow between and within organs, and (3) altering the amount of oxygen extracted from the blood. When the capacity of these compensatory mechanisms is exhausted, tissue injury occurs.

At first, compensation for the decreased oxygen content appears to be almost exclusively flow dependent. Cardiac output increases and systemic vascular resistance decreases. The increase in cardiac output initially is mainly due to reduced blood viscosity, which improves the flow properties of blood. Increased heart rate and contractility occur, but the exact mechanisms are unclear. As oxygen content decreases with progressive hemodilution, microcirculatory changes take place, leading to a recruitment of capillaries. The red cell capillary transit time increases, allowing more time from gas exchange. In addition, anemia changes the oxygen extraction ratio from the erythrocyte itself by increasing red cell 2,3-DPG and shifting the oxyhemoglobin dissociation curve to the right, allowing increased oxygen release to the tissues.
LIMITS OF ACUTE ANEMIA TOLERANCE

Animal studies have consistently demonstrated that healthy animals tolerate acute, isovolemic hemodilution to 5 g/dL without adverse consequences. Further hemodilution results in cardiac ischemia and decreased oxygen uptake. When hemoglobin levels drop below 3 g/dL there is increased lactate production, decreased left ventricular function and cardiac output, and increased risk of death. In healthy humans, the critical hemoglobin level is unknown but is likely less than 5 g/dL. Young, healthy humans have consistently been shown to tolerate acute isovolemic hemodilution without symptoms until hemoglobin reaches 7 g/dL. Below that level subjects have demonstrated increases in heart rate and subjective decreased energy levels. Subtle deficits in cognitive function begin to appear at a hemoglobin level of 6 g/dL; at 5 g/dL immediate and delayed memory are impaired, heart rate and stroke volume increase, and systemic vascular resistance decreases. In one study, two of 32 healthy adults acutely hemodiluted to 5 g/dL developed asymptomatic, reversible ST changes on electrocardiography (ECG). These nominal changes reversed immediately with transfusion to raise the hemoglobin level to 7 g/dL.

Weiskopf and colleagues made the interesting observation that the deterioration of neurocognitive function after isovolemic hemodilution from a hemoglobin of 12.7 ± 1.0 to 5.7 ± 0.3 was reversed by increasing PaO2 from around 100 to 400 mm Hg. This value is equivalent to an increase in hemoglobin concentration of roughly 3 g/dL. Similar results have been found in animal studies.

Patients who refuse blood for religious reasons have given researchers the opportunity to observe outcomes in extreme anemia. In a case-control study of 125 such patients, postoperative mortality was inversely related to the preoperative hemoglobin level, rising from 7.1% for patients with levels above 10 g/dL to 61.5% for those with levels below 6 g/dL. Viele and Weiskopf reviewed case reports of Jehovah’s Witnesses who refused blood perioperatively, and found that all patients whose deaths were attributed to anemia had a hemoglobin level 5 g/dL or lower.

Carson and colleagues, in a retrospective cohort study of surgical patients who refused blood, reported no deaths in patients, with nadir hemoglobin between 7.1 and 8.0 g/dL. Mortality for those with hemoglobin between 6.1 and 7.0 was 8.9%, and rose to 100% for patients with a hemoglobin level below 2.0 g/dL. Mortality rose sharply for those with hemoglobin less than 5.0 g/dL. Adjusted for age, cardiovascular disease and APACHE II score, the odds ratio for death for each gram decrease in hemoglobin below 7.0 was 2.1 (95% confidence interval, 1.7–2.6). The investigators reported increased morbidity for patients with hemoglobin below 7 g/dL, but this was not compared with patients with similar hemoglobin levels who received blood.

ANEMIA AND THE HEART

Brain tissue and cardiac muscle extract much more oxygen from the blood than other tissues. Cerebral oxygen extraction can increase significantly in response to acute anemia. The heart, on the other hand, is much more vulnerable to the effects of anemia. At rest the heart extracts 60% to 75% of all oxygen delivered to the coronary circulation. Consequently, if oxygen delivery is decreased due to low arterial oxygen content, the only way for the myocardium to maintain adequate oxygen delivery is by increasing coronary artery flow.

An early response to acute anemia is an increase in cardiac output, via both tachycardia and increased stroke volume, which increases myocardial oxygen
requirements. Because the myocardium relies on increased coronary blood flow as a means to compensate for an acute decrease in CaO₂, patients with coronary artery disease would seem to be at higher risks for adverse effects due to anemia. Indeed, studies in both animals and humans have suggested that individuals with cardiovascular disease are less tolerant of anemia. Animals with experimentally induced coronary stenosis demonstrate ST segment changes or locally depressed cardiac function at hemoglobin levels in the range of 7.0 to 10 g/dL, much higher than in animals with healthy coronary vessels. In humans, the clinical data are inconclusive. Patients with coronary artery disease have tolerated mild to moderate normovolemic hemodilution without showing any increase in cardiac complications or silent ischemia during ECG monitoring. However these subjects were otherwise stable patients undergoing elective hemodilution.

In a cohort study of critically ill patients, patients with cardiovascular disease had a trend toward an increased mortality when hemoglobin values were less than 9.5 g/dL (55% vs 42%, P = .09) compared with anemic patients with other diagnoses. Increasing hemoglobin values with transfusion of RBCs in these patients was associated with improved survival (odds ratio [OR] = 0.80 for each 1 g/dL increase, P = .012). However, retrospectively pooled data from 3 large observational clinical trials on 24,112 patients with acute coronary syndromes. After adjustment for baseline characteristics and nadir hematocrit, these investigators found an increased risk of death in transfused patients. Gerber, in a subgroup analysis of the TRICC trial, concluded that a restrictive RBC transfusion strategy is generally safe in most critically ill patients with cardiovascular disease, with the possible exception of patients with acute myocardial infarcts and unstable angina.

These studies were observational and retrospective, and in the Rau study the original trials from which the data was obtained were not designed to evaluate transfusion thresholds. While informative, these studies are hypothesis generating only. Anemic patients with coronary artery disease appear to be at increased risk of cardiac ischemia. However, with the exception of acute ST-elevation myocardial infarction, there are few data to show that transfusing RBCs improves outcomes. Despite this, some investigators recommend maintaining higher hemoglobin levels in patients with cardiovascular disease. Others recommend transfusing only when signs of inadequate myocardial oxygen supply start to develop.

Two trials evaluating transfusion thresholds in patients with cardiovascular disease are currently ongoing. The FOCUS trial is a multicenter randomized trial of patients with cardiovascular disease undergoing hip surgery. Patients will either have postoperative hemoglobin levels kept above 10 g/dL, or be transfused only when symptomatic from anemia. Results are expected to be published next year. The CRIT pilot trial will evaluate the effect of liberal versus restrictive transfusion strategy on outcome of anemic patients within 72 hours of myocardial infarction.

RED CELL PHYSIOLOGY

RBCs are now considered as much more than just innocuous little packets of hemoglobin. When oxygen content falls, red cells can regulate blood flow by releasing mediators like nitric oxide (NO) and ATP, which can induce local vasodilation. Hemoglobin itself may act as a tissue oxygen sensor. After a life span of 120 days, RBCs are removed from the circulation by macrophages, primarily in the spleen. This process may also occur in other microvascular networks that become obstructed by aged red cells. Various signals can induce the exposure of epitopes on RBCs that indicate senescence; these epitopes are
recognized by naturally occurring autoantibodies, initiating complement-dependent phagocytosis of aged or oxidatively stressed erythrocytes.60

Until recently, mature erythrocytes were not thought to be capable of undergoing apoptosis because the cells lack mitochondria and nuclei. However, it has now been shown that RBCs can undergo programmed cell death in response to oxidative stress. Therefore, oxidative stress due to critical illness may shorten RBC life span.60

RBCs are highly deformable, and as such are capable of passing through capillaries with diameters much smaller than their own.55,59 This flexibility is a critical factor in ensuring microvascular function as well as ensuring efficient oxygen uploading in the lungs and offloading in the tissue.59 Maintenance of red cell deformability depends on interactions between the outer plasma membrane and the underlying protein cytoskeleton.60 Increases in intracellular hemoglobin, loss of biconcave shape, decreased erythrocyte ATP, or increased intracellular Ca\(^{2+}\) can all cause decreased RBC deformability.60,61 The mechanisms are complex and only partially understood.59–62

The RBC membrane contains glycolytic enzymes, and is capable of synthesizing 2,3-DPG and ATP.56 Alterations in the RBC membrane could therefore not only change cell shape and malleability but also alter RBC biochemistry.55

In critically ill patients, especially in sepsis, RBCs undergo structural and functional changes that are similar to those in naturally aged erythrocytes55,60 RBC membrane components are modified, resulting in changed cell shape, decreased deformability,63 and increased red cell aggregation.17,55,62,64–66 These changes have been implicated in alteration of microvascular hemodynamics with a concomitant decrease in oxygen use and increased tissue ischemia.59,62

Inflammation may alter RBCs, and they in turn may affect the inflammatory response. Erythrocytes can bind opsonized immune complexes, restricting immune complex uptake and degranulation by macrophages. Oxidatively damaged RBCs can increase tumor necrosis factor (TNF)-\(\alpha\) and interleukin (IL)-10 production by monocytes. Thus, inflammation appears to cause functional and structural changes in RBCs, but these altered RBCs can themselves modulate the immune response.60

**BLOOD AND THE MICROCIRCULATION**

The microcirculation is where oxygen diffusion takes place, and consists of all vessels smaller than 100 \(\mu\)m, including arterioles, capillaries, and venules.27,55 Local microcirculatory control mechanisms, although poorly understood, are important in the maintenance and restoration of tissue oxygenation.27

Blood viscosity is an important regulator of blood flow.18 Blood viscosity in turn depends on the concentration of cells, the ability of cells to aggregate and deform, and on plasma viscosity.61 Of these, the hematocrit is the most important.67 Blood viscosity is inversely related to flow, so that for a given vessel diameter an increase in hematocrit will reduce blood flow exponentially.61 Very low hemoglobin concentrations impair the delivery of oxygen to tissues; paradoxically, a moderate decrease in hematocrit might improve oxygen transport by lowering blood viscosity.33,60,61,68

However, maintenance of blood viscosity is important in maintaining microvascular perfusion.69 Increasing blood viscosity increases shear stress, which increases endothelial release of nitric oxide, resulting in local vasodilation. Experimentally, restoration of blood rheological properties improves resuscitation independently of any improvement in oxygen-carrying capacity.70 Beneficial effects of blood transfusions may therefore be in part due to the increase or restoration of blood viscosity.59
THE STORAGE LESION

In the United States the average age of RBCs transfused in the critically ill is 21 days.\textsuperscript{2,10,71,72} When stored, RBCs undergo several well-documented biologic and mechanical changes known as the “storage lesion.” In 1997 Purdy and colleagues\textsuperscript{73} first reported a correlation between increased age of transfused blood and increased mortality. Despite millions of transfusions since then, the clinical consequences of the storage lesion remain uncertain and controversial.\textsuperscript{17,74}

A red cell normally contains 20 to 25 $\mu$mol of 2,3-DPG, which shifts the oxyhemoglobin dissociation curve to the right, allowing hemoglobin to release oxygen.\textsuperscript{17,59} Erythrocyte 2,3-DPG levels begin to decline shortly after collection, and are near zero within 48 to 72 hours.\textsuperscript{13,18,25,27,59,75} Therefore transfused cells are able to oxygenate well as they pass through the lungs, but in principle are unable to release oxygen to the tissue unless the tissue oxygen tension is very low.\textsuperscript{8,10,18} Within several hours after transfusion 2,3-DPG levels begin to increase, and gradually return to normal by 24 hours after transfusion.\textsuperscript{8,27,76}

Erythrocyte deformability is an energy-dependent process,\textsuperscript{59} sustained by intracellular catabolism of glucose and generation of high-energy ATP.\textsuperscript{17} RBC ATP levels drop with storage,\textsuperscript{74,77} but it is not clear at what level RBC deformability is affected. Within the first 2 weeks after collection, erythrocytes begin to change shape and lose flexibility.\textsuperscript{78} The RBC membrane loses proteins and phospholipids, and cells change from supple biconcave discs to stiff echinocytes and spherocytes.\textsuperscript{17} When viewed under an electron microscope, these abnormal cells account for as much as 80% of the cell population at 3 weeks, and more than 95% by 35 days.\textsuperscript{8,79} Such cells, when transfused, become trapped in the microvasculature\textsuperscript{80,81} or are eliminated by the reticuloendothelial system.

In the normal erythrocyte, ATP stimulates NO production by endothelial cells. Transfusion of ATP-depleted RBCs may lead to decreased NO production and increased vascular resistance.\textsuperscript{59} Similarly, erythrocyte S-nitrosohemoglobin concentrations have been noted to decline rapidly after red cell storage, decreasing the ability to locally control vasodilation.\textsuperscript{82}

Within several hours of transfusion, ATP levels in viable cells are restored by uptake of adenosine from the plasma.\textsuperscript{27} With prolonged storage time, however, a high percentage of cells will be irreversibly deformed and no longer viable.\textsuperscript{17,79}

These misshapen cells exhibit increased osmotic fragility, releasing free hemoglobin into the plasma.\textsuperscript{18,19,27,74,79} Free hemoglobin levels in stored cells increase significantly over time.\textsuperscript{83} Free hemoglobin scavenges NO, even in very low concentrations, resulting in local and systemic vasoconstriction.\textsuperscript{8,18,27,84}

Hemoglobin is a profound oxidizer and is highly toxic to endothelial cells. Endothelial cells pinocytose free hemoglobin, which leads to endothelial cell dysfunction exhibited as reperfusion injury and oxidative stress. Such cells, rather than being naturally anti-inflammatory, become proinflammatory and highly thrombotic.\textsuperscript{18}

RBC storage increases RBC-endothelial interactions,\textsuperscript{85} which are further increased by endotoxins and inflammatory cytokines.\textsuperscript{17,86} A unique interaction between the RBC storage lesion and an injured microcirculation in sepsis might explain the inability to demonstrate improved tissue oxygenation after RBC transfusion in sepsis.\textsuperscript{7}

Stored erythrocytes exhibit increased aggregation and adhesiveness,\textsuperscript{74,79} and this increases with storage time.\textsuperscript{18,79} The mechanism for this is unclear. Sialic acid maintains a negative charge on the cell surface, and it has been hypothesized that decreased membrane sialic acid contributes to the aggregation.\textsuperscript{74,79}
Storage Medium

In addition to alterations in the RBCs themselves, changes to the storage medium also occur. With time there is a progressive decrease in pH, increase in potassium, accumulation of cytokines (IL-1, IL-8, TNF)\(^72\) and accumulation of other bioreactive substances such as histamine, soluble lipids, lysophosphatidylcholine.\(^74\)

Studies

Together, the well-recognized changes to stored RBCs could decrease the benefit of transfusion, and possibly cause harm. Several animal studies have evaluated the clinical effect of red cell storage, with mixed results.\(^8,77,87,88\)

Observational and retrospective studies in humans have shown age of transfused red cells to be associated with splanchnic ischemia,\(^25\) increased risk of infection,\(^89–93\) multiorgan failure,\(^94\) increased hospital or ICU length of stay,\(^95,96\) and mortality.\(^73,93,96\) Weinberg and colleagues,\(^97\) in their recently conducted retrospective cohort study to evaluate the association between the age of transfused blood and mortality among trauma patients, found that large volumes of transfused blood were associated with increased mortality and that the effect was potentiated with older blood. However, a large observational trial by van de Watering and colleagues\(^98\) found no association between a prolonged storage time and increased mortality or longer stay in the ICU.

A recent literature review was inconclusive. The majority of studies analyzed were single-center with small sample size, and patient populations were heterogeneous, making it difficult to compare studies.\(^74\)

There are limited randomized controlled trials evaluating the clinical effects of red cell storage. In mechanically ventilated, critically ill patients, Walsh and colleagues\(^99\) compared packed RBCs of age 5 days or less versus 20 days or older, and found no difference in gastric pH or in global indices of tissue oxygenation. In a pilot trial to evaluate the feasibility of maintaining a supply of fresh blood, Hebert and colleagues\(^72\) found no differences in major clinical outcomes between fresh and older blood.

At the microvascular level, Kiraly and colleagues,\(^100\) using NIRS, found that transfusing old blood into trauma patients worsened microvascular oxygenation. Weiskopf and colleagues\(^101\) showed that transfusion of “fresh” (mean age 3.5 ± 0.4 hours) and “old” (mean age 23 ± days) autologous erythrocytes to increase the hemoglobin level from 5 to 7 g/dL equally reversed neuropsychological deficits in acutely hemodiluted healthy volunteers.

Although theoretically and potentially harmful, it is difficult to determine from the currently available published data whether there is a relationship between the age of transfused RBCs and outcome in critically ill patients.\(^74\)

TRANSFUSION-RELATED IMMUNOMODULATION

Although not definitively proven in randomized controlled trials, allogeneic blood transfusions have long been known to have immunosuppressive effects. Transfusion-related immunomodulation, or TRIM, refers to the proinflammatory and immunomodulatory laboratory findings and clinical effects of blood transfusion.\(^102,103\)

In 1973 Opelz and colleagues showed improved survival in renal allograft recipients who had received blood. In the early 1980s, renal transplant recipients were often transfused prior to transplant to decrease rejection.\(^104\) This practice fell out of favor with the recognition of AIDS as a significant risk of transfusion.\(^102\)
Although TRIM is a widely acknowledged effect of blood transfusion, the exact clinical consequences remain uncertain. Observational studies show a strong correlation between blood transfusion and nosocomial infection, but a causal effect has not yet been proven. Other effects seen in observational studies include a reduced risk of Crohn’s recurrence, decreased risk of spontaneous abortion in mothers who share human leukocyte antigen (HLA) antigens with the father, increased risk of activation of latent cytomegalovirus or human immunodeficiency virus infection, increased risk of cancer recurrence, and increased risk of multiple organ failure and all-cause mortality.

**Infection**

Several observational studies in humans have linked packed red cell transfusion with nosocomial infection. In a meta-analysis of such trials by Hill and colleagues, the overall OR for transfusion and postoperative bacterial infection was 3.45 (range 1.43–15.15).

Recently, Taylor and colleagues performed a large prospective, observational, cohort study of more than 2000 critically ill patients. Just over 21% received transfusions, with a mean pretransfusion hemoglobin level of 7.6 g/dL. The rate of nosocomial infection in transfused patients was 14.3%, significantly higher than the nosocomial infection rate of 5.8% in the nontransfused group (P < .0001). After multivariate logistic regression analysis of possible risk factors including patient age, the total units of RBCs transfused, or the maximum age of RBC units transfused, the number of RBC units transfused was the only significant, independent risk factor for nosocomial infection (OR 1.097, P = .005). That is, for every unit of packed cells transfused, the risk of infection increased 9.7%. Although the link appears strong, a definitive causal relationship between red cell transfusion and infection has not yet been established.

**Cancer**

Animal data and observational studies have linked transfusion of RBCs to increased risk of cancer recurrence. To date there have been 3 randomized controlled trials of allogeneic blood transfusion and cancer recurrence, all in colorectal cancer patients after curative surgical resection, and the results are inconclusive.

Heiss and colleagues found significantly increased recurrent malignancy in colorectal cancer patients who received blood perioperatively at the time of resection. On the other hand, neither Busch and colleagues nor Houbiers and Brand noted increased cancer recurrence with transfusion in similar patient populations.

Vamvakas and Blajchman have speculated that the negative results in the aforementioned studies are at least in part because colorectal cancer is not sufficiently antigenic to render an impairment of host immunity capable of facilitating tumor growth. To date, no randomized controlled trial has enrolled patients with tumors for which the immune response plays a major role, such as skin tumors and certain virus-induced tumors like Kaposi sarcoma and certain lymphomas.

**Leukocytes**

Which component or components of blood mediates TRIM, and by what mechanism, are unknown. Allogeneic blood transfusion has been shown to cause decreased helper T-cell count, decreased helper/suppressor T-lymphocyte ratio, decreased lymphocyte response to mitogens, decreased natural killer cell function, reduction in delayed-type hypersensitivity, defective antigen presentation, suppression of lymphocyte blastogenesis, decreased cytokine (IL-2, interferon-γ) production, decreased monocyte/macrophage phagocytic function, and increased production of anti-idiotypic and anticonnotypic antibodies.
TRIM is thought by many to be mediated mainly by donor white blood cells.116 Most transfusions currently administered in North America and Western Europe are leukoreduced red cells.117 Leukoreduction removes approximately 99% of the white blood cells present in whole blood, which still leaves approximately $10^6 - 10^9$ white cells per unit.18,118

Animal models suggest leukocytes as probable causative agents of immunomodulation, but this has not been proven in humans.103,104 Donor leukocytes may either directly downregulate the recipient’s immune function, or indirectly mediate TRIM effects by releasing soluble mediators into the supernatant fluid of RBCs during storage.109

Leukocyte-derived soluble mediators are contained in intracellular white blood cell granules, and are released in a time-dependent manner as the leukocytes deteriorate during storage. Concentrations of histamine, eosinophil cationic protein, eosinophil protein X, myeloperoxidase, and plasminogen activator inhibitor-1 increase 3- to 25-fold in the supernatant fluid of packed reds cell between days 0 and 35 of storage.109 Packed red cell supernatant induces regulatory T cells in recipients, but this is not altered by leukoreduction or prolonged storage. This induction seems to be independent of cytokines and is attenuated with washed red cells, implicating the plasma fraction.105

Frietsch and colleagues106 recently conducted a multicenter, double-blind, randomized controlled trial comparing prestorage leukoreduced autologous blood to nonleukoreduced autologous blood in 1089 patients undergoing elective hip surgery. There was no difference between the 2 groups in postoperative infection rate or hospital length of stay, which argues against leukocyte-released biologic modifiers as a cause of the immunomodulatory effects of blood transfusion.

With the exception of cardiac surgery, the benefit of leukoreduction on overall mortality and on infectious complications is highly debated.116 In 1998, van de Watering and colleagues105 performed a randomized controlled trial comparing cardiac surgery patients receiving standard versus leukocyte-reduced allogeneic packed red cells, and found an association between the leukocyte-containing packed red cells and mortality from all causes. These results were confirmed in a repeat trial by the same investigators,119 and in a similar randomized controlled trial by Boshkov and colleagues.120

A recent literature review103 analyzed 19 randomized controlled trials on the effect of allogeneic leukocytes in transfusions. With the exception of the aforementioned cardiac surgery trials, the results were inconclusive. The mechanism of increased complications with cardiac surgery is uncertain, but the investigators suggest that leukocyte-containing transfusions during and after cardiac surgery add a second insult to the cardiopulmonary bypass procedure induced systemic inflammatory response.

Hebert and colleagues121 retrospectively examined outcomes of critically ill patients in Canada before and after implementation of universal leukoreduction. These investigators found a small, but statistically significant, decrease in all-cause mortality (6.19% vs 7.03%; $P = .04$) following universal leukoreduction. There was no significant decrease in infections.

Fergusson and colleagues122 retrospectively compared outcomes of premature infants in Canada before and after implementation of universal leukoreduction. There was no difference in mortality or in the risk of bacteremia between the 2 study periods, but there was a reduction in bronchopulmonary dysplasia, retinopathy of prematurity, and necrotizing enterocolitis. This result would be consistent with a proinflammatory effect of allogeneic leukocytes.109 Englehart and colleagues123
recently performed a retrospective cohort analysis of trauma patients at a large level I trauma center that used only leukoreduced blood between 2002 and 2003. Before 2002, and after 2003, nonleukoreduced blood was standard. Englehart’s group found no improvement in outcome in trauma patients receiving leukoreduced blood.

Transfusion-Related Lung Injury

Transfusion-related lung injury, or TRALI, is an increasingly recognized risk of blood transfusion. The estimated prevalence of TRALI is 1 in 5000 units of transfused RBCs. TRALI is characterized by acute onset of severe hypoxemia, bilateral non-cardiogenic pulmonary edema, tachycardia, hypotension, and fever. With supportive care, most patients recover within 48 to 96 hours, but mortality can be as high as 25%.

TRALI remains underrecognized, in part due to the lack of a standardized diagnostic criteria, possible confusion with other causes of acute respiratory failure, and lack of a definitive diagnostic test. The National Heart, Lung and Blood Institute working group on TRALI recently presented a common definition of TRALI, and advised consideration of the diagnosis in any patient that develops acute lung injury within 6 hours of transfusion, with no preexisting acute lung injury before transfusion. Alternative causes of acute lung injury do not preclude a diagnosis of TRALI.

The pathophysiology of TRALI is still being elucidated and is likely multifactorial, involving both priming of recipient neutrophils by underlying condition and biologic response modifiers with neutrophil-priming activity in the transfused blood product. Most cases occur with blood products containing at least 50 mL of plasma, and TRALI is 17 times more likely with cellular blood products than with fresh frozen plasma. Anti-polymorphonuclear and anti-HLA class I antibodies in donor plasma have been strongly implicated in TRALI, and exclusion of female donor plasma by the UK National Blood Services reduced the onset of acute lung injury from 36% to 21% (P = .04) in patients receiving multiple transfusions while undergoing elective repair of a ruptured abdominal aortic aneurysm.

CLINICAL TRIALS

Despite the prevalent use of blood transfusion as a therapy, there have been surprisingly few randomized, controlled trials evaluating its effectiveness. Hebert’s TRICC trial, published in 1999, was the sentinel study that changed transfusion practice. Hebert and colleagues enrolled 838 critically ill patients in a multicenter, randomized controlled trial comparing a restrictive transfusion strategy (maintaining hemoglobin level between 7 and 9 g/dL) to a more liberal one (maintaining hemoglobin level between 10 and 12 g/dL). Not only was the restrictive strategy shown to be safe, but for younger (age <55 years) and less ill patients (APACHE II < 20), mortality was lower in the restrictive group.

Based largely on the trial by Rivers and colleagues of early goal-directed therapy (EGDT) in sepsis, current guidelines recommend a transfusion threshold of 10 g/dL in septic patients who are apparently oxygen dependent based on central venous oxygen saturation (ScvO2). As pointed out by Jha and Gutierrez, almost 65% of the EGDT subjects received transfusions, despite the fact that basal hematocrit was almost 35%. The intervention group received a large volume of intravenous fluids, resulting in greater hemodilution. According to the Fick principle, SvO2 varies directly with arterial oxygen saturation, cardiac output, and hemoglobin concentration, and inversely with oxygen consumption. Hemodilution-induced decreases in hemoglobin
concentration would depress SvO₂, regardless of whether the patients had entered an oxygen supply–dependent state. As discussed earlier in this article, transfusion of red cells could improve oxygen delivery systemically, without actually improving oxygen uptake by the tissues. Furthermore, central venous saturation has not been thoroughly validated as a measure of adequacy of DO₂ but was used in this study as a central end point for resuscitation.¹⁰

Numerous trials have demonstrated an association between red cell transfusions and infection,⁷¹,¹⁰⁷,¹¹⁰,¹³⁰ multiple organ failure,⁹⁴,¹³¹ increased ICU and hospital length of stay,⁷¹,⁹⁵,¹³² and mortality.⁷¹,⁷³,⁹⁷,¹³²–¹³⁴ Unfortunately most of these studies are observational, and cause and effect can only be proven by appropriately powered randomized controlled trials.¹⁸ There are very few randomized controlled trials, other than the TRICC trial, comparing clinical outcomes between transfused and nontransfused patients. However, the sheer bulk of the data implicating transfusions in worsened clinical outcomes is compelling.

The primary goal of RBC transfusion is to improve oxygen delivery to tissues that are presumably oxygen dependent, or near their critical oxygen delivery threshold. Several clinical trials have examined the effect of RBC transfusion on oxygen consumption. In one of the few randomized controlled trials in septic patients, Fernandes and colleagues¹³⁵ determined that RBC transfusion did not increase oxygen use, whether indirectly calculated by the Fick method or directly measured by calorimetry. Shah and colleagues¹³⁶ found similar results in trauma patients. Suttner and colleagues¹³⁷ measured skeletal muscle oxygen tension in postoperative cardiac surgery patients. Transfusion improved systemic oxygen delivery, but had no effect on tissue oxygen tension. Conversely, inspiration of 100% oxygen improved both oxygen delivery and tissue oxygen tension. Walsh and colleagues⁹⁹ recently conducted a randomized trial to compare fresh with old blood. Although these investigators found no difference in oxygen uptake between the two, there was also no statistically or clinically significant improvement in any other oxygenation index for either group. In other words, even though the patients’ attending physicians felt they would benefit from blood transfusion, objectively they showed no improvement after transfusion. Kiraly and colleagues,¹⁰⁰ using NIRS to measure tissue oxygen saturation in trauma patients, found no improvement with transfusion of fresh blood, and a deterioration of tissue oxygen saturation with blood older than 21 days.

**SUMMARY**

The concept of a universal transfusion threshold is very appealing. For years physicians used the “10/30” rule, but it is clear that this is no longer a clinically useful value. Most critically ill patients can tolerate much lower hemoglobin levels without adverse effects, but a safe threshold above which red cell transfusion is clearly unnecessary has not been established.³⁷,³⁸ The ideal would be to identify a “critical hemoglobin concentration”—the point at which compensatory mechanisms for anemia have been maximized and further reduction in hemoglobin would result in compromised cellular metabolism—in individual patients.²¹ At present clinicians have neither the technology to detect DO₂(crit) nor the knowledge of how near to that level an individual patient might be.¹⁸

The goal of RBC transfusion is to improve tissue oxygenation; however, transfused red cells may increase oxygen delivery to the tissues without a corresponding increase in oxygen consumption.¹¹⁸ First, most anemic patients do not have tissue hypoxia unless they also have acute circulatory failure.¹³ Second, red cells may not be effective
in their role as oxygen transporters, and they are now believed to pose additional risks for critically ill patients.71

The best available evidence does not support the use of a single criterion for transfusion. In the absence of acute cardiac ischemia or acute bleeding, hemoglobin levels of 7 to 9 g/dL are well tolerated in the critically ill.118 The decision to transfuse should be based on thoughtful clinical judgment, on an individual basis.135,138

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