Anemia in Critical Illness

Insights into Etiology, Consequences, and Management

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Anemia is common in the intensive care unit, and may be associated with adverse consequences. However, current options for correcting anemia are not without problems and presently lack convincing efficacy for improving survival in critically ill patients. In this article we review normal red blood cell physiology; etiologies of anemia in the intensive care unit; its association with adverse outcomes; and the risks, benefits, and efficacy of various management strategies, including blood transfusion, erythropoietin, blood substitutes, iron therapy, and minimization of diagnostic phlebotomy.

Keywords: anemia; red blood cell; transfusion; erythropoietin; critical illness

Anemia is highly prevalent in critically ill and injured patients. Approximately two-thirds present with a hemoglobin concentration less than 12 g/dl on admission, and 97% become anemic by Day 8 (1–3). Optimal management of the anemia of critical illness is an area of much controversy and ongoing research. In this article, we first describe normal red blood cell (RBC) physiology and the etiologies and effects of anemia, and then review potential management strategies.

RED BLOOD CELL FUNCTION

The efficient blood transport of oxygen and carbon dioxide is critically dependent on the O_2, CO_2, and H^+ binding properties of hemoglobin (Hb). These are facilitated by the enzyme carbonic anhydrase, RBC-specific membrane and cytoplasmic proteins, and by the unique intracellular RBC environment, including the modulation of Hb–O_2 affinity by 2,3-diphosphoglycerate (2,3-DPG) and maintenance of redox state stability. Packaging the majority of the blood's O_2 and CO_2 transport capacity in the RBC reduces the effective viscosity of blood by two-thirds in comparison with a cell-free medium of equivalent transport capacity and prevents loss of hemoglobin, carbonic anhydrase, and 2,3-DPG via glomerular filtration. RBCs also have potent antioxidant capacity, enhance hemostasis by directing platelets toward the vessel wall, minimize hemoglobin–nitric oxide (NO) scavenging by sequestrating Hb away from direct contact with the endothelium of resistance arterioles, and play an important role in microcirculatory vasoregulation.

RBC rheology contributes to vasoregulation, particularly at the microvascular level (4). In addition to tissue and endothelial cell contributions to vascular tone mediated in part by tissue oxygenation, rheology has an influence on vascular tone by altering wall shear stress and nitric oxide generation, as well as by homogenizing flow distribution at capillary branch points. Local blood flow and metabolic demand are matched by three mechanisms involving RBCs and hemoglobin. First, bioactive NO is produced in proportion to the concentration of deoxyhemoglobin acting as a nitrite reductase (5). Second, NO is bound by oxyhemoglobin in the lungs and released by deoxyhemoglobin in the tissues, mediated by reversible allosteric S-nitrosylation of β-chain cysteine-93 in hemoglobin (6). Third, mechanical deformation of RBCs and desaturation of hemoglobin initiates vasodilation by release of ATP that binds to endothelial cell purinergic receptors and stimulates NO synthesis (7). The redundancy of these mechanisms highlights the likely critical contribution of healthy RBCs to active vasoregulation. However, this function may be compromised both by anemia and by pathological RBC changes occurring in critical illness and during storage of allogeneic blood.

REGULATION OF RED BLOOD CELL MASS

With a life span of only 120 days, caused by accumulative radical oxygen species (ROS)–mediated damage and age-related loss of intrinsic antioxidant defenses (8), there must be a constant production of new RBCs. The essential factors for erythropoiesis include iron, zinc, folate, and vitamin B_{12}, under the influence of erythropoietin (EPO), thyroxine, androgens, cortisol, and catecholamines. RBC formation occurs at a basal rate of 15–20 ml/day under steady-state conditions, and upward of 200 ml/day after hemolysis or heavy blood loss in iron-replete healthy persons (9). Normal RBC aging leads to changes in membrane characteristics (reduced fluidity and deformability), loss of volume and surface area, increased cell density and viscosity, and deleterious alterations in the intracellular milieu (decreased ATP and 2,3-DPG, lowered hexokinase and glucose-6-phosphate dehydrogenase activity). These lead to a fall in cellular energy level, increased Hb–oxygen affinity, reduced repair of oxidant injury, and diminished ability of cells to deform normally during microvascular transit (10). These changes also mark RBCs for removal by the spleen and reticuloendothelial system (RES). Alterations in rheology with normal aging may occur even sooner in the life span of the RBC in critically ill patients, and have been shown to be associated with poor outcomes (11).

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Other determinants of RBC survival include the premature death of mature RBCs (eryptosis), and removal of RBCs just released from the marrow (neocytolysis). Eryptosis is thought to be partially triggered by excessive oxidant RBC injury, among other stressors, and is inhibited by EPO, which thus extends the life span of circulating RBCs (12). This apoptosis-like process is characterized by a cascade of biochemical and biomechanical changes, leading to cell shrinkage, dysregulation of normal membrane asymmetry with exposure of normally sequestered phosphatidylserine on the outer membrane leaflet, and the formation of membrane blebs and microparticles. Phosphatidylserine marks cells for engulfment by macrophages and may carry important downstream effects related to inflammation, coagulation, cell signaling, and/or immune modulation. Conversely, excessive eryptosis may lead to the development of anemia (13). Neocytolysis is the process of selectively removing young circulating RBCs, initiated by a sudden fall in EPO levels (14). This phenomenon was first noted in the study of RBC mass reduction that occurs during spaceflight (microgravity) and after descent from high altitude; as both processes develop too rapidly to be accounted for solely by reduced erythropoiesis. Eryptosis and neocytolysis, negatively regulated by EPO and acting at different points in the life span of the RBC, provide flexibility and fine control in regulation of total RBC mass.

ETIOLOGY OF ANEMIA IN CRITICAL ILLNESS

In critical illness and injury, anemia results from two fundamental processes: a shortened RBC circulatory life span and diminished RBC production. Causes of shortened life span include hemolysis, phlebotomy losses, oozing at injury sites, invasive procedures, and gastrointestinal bleeding. Diagnostic phlebotomy in the critically ill represents a mean daily loss of 40 to 70 ml of blood, exceeding the normal healthy replacement rate (1, 15). Of blood sent for analysis, less than 2% is actually assayed with modern laboratory instrumentation. This phenomenon, termed the "anemia of chronic investigation" (16), accounts for 30% of required blood transfusions (15). The impaired mucosal integrity of the gastrointestinal (GI) tract is also a source of on-going occult blood loss (17). Cook and colleagues showed that clinically important bleeding from stress gastritis occurs in 3% of ICU patients, with the main risk factors being mechanical ventilation, nutritional deficiencies, acute renal failure, and prophylactic or therapeutic anticoagulation (18).

Diminished RBC production is due to nutritional deficiencies and the "anemia of inflammation." In one study, 9% of ICU patients were iron deficient, with an additional 2% each to B_{12} and folate deficiency (19).

The "anemia of inflammation" collectively refers to inflammatory processes leading to impaired RBC proliferation, iron metabolism, EPO production, and signaling. It is, in part, thought to be a broad-based evolutionary response to sequester and deny iron to invading micro-organisms. Numerous proinflammatory cytokines, including IL-1, IL-6, and tumor necrosis factor (TNF)-α, impair iron homeostasis and normal RES functioning, and decrease regulatory feedback between body iron needs and intestinal iron absorption (20). Hepcidin, an iron regulatory protein that is up-regulated in inflammatory conditions and suppressed by EPO, decreases duodenal iron absorption and blocks iron release from macrophages. This limits iron availability for erythroid progenitor cells and impairs heme biosynthesis, leading to iron-restricted erythropoiesis (21). During systemic infection there is up-regulation of the IL-6–hepcidin axis, which in part may be responsible for low serum iron levels observed in inflammation (22). The minimal response by the bone marrow is possibly caused by reduced transcription of the EPO gene by inflammatory mediators such as IL-1, TNF-α, and transforming growth factor (TGF)-β. These same inflammatory cytokines also inhibit RBC production through direct interactions with erythroid progenitor cells (23). Finally, in the setting of shock, these effects might be magnified by vasopressor agents such as norepinephrine or phenylephrine, which at high concentrations directly inhibit hematopoietic precursor maturation (24).

Erythropoiesis is tightly regulated by EPO, the levels of which are normally increased with anemia. However, during critical illness, circulating EPO concentrations fall quickly and remain inappropriately low because of a combination of decreased renal function and proinflammatory cytokine inhibition of EPO production (25, 26). In addition to suppressed RBC production, a sudden and continued drop in EPO production with the onset of any acute inflammatory condition may promote neocytolysis and eryptosis as discussed earlier. Furthermore, the response to EPO is blunted through down-regulation of EPO receptors, limiting the availability of iron for cell proliferation and hemoglobin synthesis (21, 26, 27). Last, some very commonly used drugs in the ICU, such as angiotensin-converting enzyme inhibitors, angiotensin receptor blockers, calcium channel blockers, theophylline, and β-adrenergic blockers, also suppress the normal renal EPO release in response to hypoxemia and anemia (28–30).

PHYSIOLOGICAL EFFECTS OF ANEMIA

Acute or chronic anemia requires compensatory responses that place an extra burden on critically ill patients, many of whom have preexisting cardiopulmonary disease. Acute isovolemic reduction in hemoglobin concentration to as low as 5 g/dl in resting healthy humans leads to progressive increases in heart rate, stroke volume, oxygen extraction, and cardiac index (31), with no evidence of tissue hypoxia. Even more severe anemia may be tolerable with chronicity due to changes at the cellular level driven by transcription of genes enhancing hypoxic survival, such as those regulated by hypoxia-inducible factor (HIF) (32). In patients with chronic obstructive pulmonary disease (COPD), there is higher minute ventilation with anemia (33), and conversely, less ventilation with polycythemia in healthy persons during exercise (34). The extent to which these impressive compensatory changes can occur in critically ill and injured patients is unknown.

ASSOCIATION BETWEEN ANEMIA AND ADVERSE OUTCOMES

A strong association exists between anemia and poor patient outcomes across numerous chronic diseases. In more than 12,000 older adults with normal renal function, anemia was associated with increased risk of mortality (hazard ratio, 4.29) and hospitalization (hazard ratio, 2.16), after adjusting for age, sex, diabetes, and chronic disease score (35). Analyses of the medical arm of the National Emphysema Treatment Trial (36) and of patients in the French ANTADIR database with severe O_2-requiring COPD (37) identified anemia as an independent predictor of death. The effect of anemia on mortality in patients with COPD is greater with increasing anemia severity (38). Many trials have shown an association between anemia and adverse outcomes in congestive heart failure (39), acute myocardial infarction (40), and chronic kidney disease (41).

ICU anemia is also associated with adverse outcomes, including failure of liberation from mechanical ventilation (42), type 2 myocardial infarction (injury due to imbalance in oxygen supply and demand) (43), and increased risk of death (44, 45). In a cohort of 91 patients recovering from acute respiratory failure, those with a hemoglobin concentration less than 10 g/dl were

more than five times as likely to require reintubation after an initial successful spontaneous breathing trial and extubation (42). Among 222 patients with COPD requiring invasive mechanical ventilation, the adjusted risk of death within 90 days of admission for anemic patients (hemoglobin < 12 g/dl) was 2.6 times that of those with normal values (12–15 g/dl) (44). Moreover, in 300 surgical patients who declined blood transfusion for religious reasons, the adjusted odds of death within 30 days of surgery was 2.5 for each gram decrease in hemoglobin below 8 g/dl (45). However, there were no deaths among the small number of patients with levels of 7.1 to 8.0 g/dl. Anemia also leads to overestimation of serum glucose by point-of-care glucometers, creating a risk of hypoglycemia if these values are used to dose insulin (46). The subtle reversible cognitive dysfunction found during severe acute anemia in healthy subjects (47) is hypothesis-generating regarding the relationship of anemia and delirium in the ICU.

Anemia often persists long after ICU discharge. In one study, 53% of patients with anemia at ICU discharge were still anemic 6 months later (48). Although not specifically studied in ICU survivors, extrapolation from the literature on anemia in various chronic illnesses described above suggests that persistent anemia after critical illness may carry important long-term consequences.

In all of these studies, it is difficult to ascertain whether anemia is an independent predictor of poor outcomes or merely a marker of more severe underlying disease not captured in chosen parameters of disease severity. Nonetheless, the stresses of compensatory cardiopulmonary responses to anemia discussed earlier create a plausible mechanism for a causal relationship.

Prospective trials in ICU patients, discussed in more detail below, have failed to show a survival benefit from a liberal transfusion strategy or treatment with erythropoietin (49, 50). These results do not imply that anemia is harmless, but they do raise concerns regarding potential detrimental outcomes linked to RBC transfusions and erythropoietin.

MANAGEMENT OF ANEMIA IN CRITICAL ILLNESS

Blood Transfusions

Anemia in critical illness has traditionally been treated with RBC transfusions. More than one-third of all ICU patients receive transfusions and more than 70% when ICU stay exceeds 1 week (1, 2, 51). In the United States, 14.7 million units of allogeneic RBCs were transfused in 2006, at a mean procurement cost of $214 per unit, which does not include costs of labor, further laboratory testing, and a myriad of adverse reactions (52). Transfusion practice is an area of controversy in critical care medicine, as discussed below.

Potential benefits. The primary goal of transfusion in the volume-replete nonhemorrhaging patient is to improve tissue oxygen delivery and carbon dioxide removal. As oxygen delivery is largely determined by cardiac output, hemoglobin concentration, and oxygen saturation, changes in hemoglobin will have a significant effect. However, studies of blood transfusions in acute respiratory distress syndrome (ARDS), sepsis, and trauma have not shown any improvement in oxygen uptake (53–56). This may be due to partially reversible biochemical and structural changes in stored blood, collectively termed the RBC storage lesion, which may inhibit oxygen unloading, normal capillary flux, and tissue oxygenation (57, 58), particularly in the first 12–24 hours after transfusion, as for example in the regeneration of 2,3-DPG (59).

Potential harms. Potential adverse effects from allogeneic blood include transfusion reactions, transfusion-transmitted infections, acute lung injury (ALI) and transfusion-related acute lung injury (TRALI), transfusion-associated circulatory overload (TACO), and transfusion-related immunomodulation (TRIM), potentially leading to increased risks of nosocomial infection and death. With modern blood banking, the risk for transfusion-transmitted viral or bacterial infection is extremely low (60). TRALI, TACO, and TRIM, although infrequent, have substantial cost and morbidity (reviewed in References 61 and 62). Moreover, transfused RBCs may not immediately improve tissue oxygenation owing to their poor flow characteristics, sludging in capillaries, high O_2 affinity, vasoconstriction due to free hemoglobin and microparticles, and, in some cases, high carboxyhemoglobin levels.

Many observational studies find associations between transfusions and poor clinical outcomes in high-risk hospitalized patients. A systematic review detailed these studies, and determined a pooled odds ratio of 1.8 for nosocomial infection, 2.5 for ARDS, and 1.7 for mortality (63). In general ICU patients, the two largest trials merit individual reference. Both the 2002 ABC trial (1) of 3,534 patients in western European ICUs, and the 2004 CRIT study (2) of 4,892 patients in U.S. ICUs found blood transfusions to be an independent predictor of death. A more recent large retrospective analysis of more than 14,000 trauma patients also found an association between blood transfusions and ARDS (adjusted odds ratio, 2.5 in patients receiving more than 5 units of packed RBCs) (64). The 2008 SOAP trial (51), which used the same study protocol as the ABC trial, showed no association between transfusion and mortality. Mean pretransfusion hemoglobin was not given, so it is not possible to comment on the potential role of a more restrictive transfusion practice in explaining the difference.

The adverse outcomes associated with RBC transfusion may be, in part, mediated by the changes that occur during storage, as discussed below.

Effect of RBC storage duration. The U.S. Food and Drug Administration (FDA) mandates a maximal storage period for RBC units of 42 days, based on a requirement for sufficient cellular integrity to ensure persistence in the circulation of more than 75% of transfused RBCs 24 hours after transfusion. For ICU patients, the mean storage time before transfusion in the United States ranges from 16 to 21 days (2, 52). Many potentially deleterious changes occur during preservation and storage. These include decreased concentrations of ATP, 2,3-DPG, and *S*-nitrosylhemoglobin (65); accumulation of proinflammatory cytokines; release of hemoglobin and red cell arginase; accumulation of RBC membrane microparticles (66); and decreased RBC membrane inactivation of cytokines by Duffy antigen (67). The hypothesized impact of these changes includes potent NO scavenging and vasoconstriction, loss of normal RBC-mediated vasoregulation, and immunosuppression. The clinical consequences of RBC storage duration are difficult to study, partly because there is no consensus definition of storage duration, for example, mean age of all units or age of the oldest unit. Although a large number of studies suggest that extended storage time is associated with infections, multiple organ failure, and death, other studies show no difference in outcomes (reviewed in Reference 68). In a more recent retrospective cohort study of more than 350,000 patients in Sweden and Denmark, Edgren and colleagues found a 5% excess mortality in recipients of RBCs stored for 30 days or more, but the authors believe this risk to be inflated by residual confounding (69). Regardless, it should be emphasized that the analysis by Edgren and colleagues was vastly dominated by non-ICU patients, leaving open the possibility of increased risk in the ICU. Animal studies (70, 71) provide new insight into potential mechanisms of harm with transfusion of blood after prolonged storage. Large randomized clinical trials have been designed to address this issue,

including the Age of BLood Evaluation (ABLE) study of ICU patients in Canada (ISRCTN 44878718), and the Red Cell Storage Duration (RECESS) study of cardiac surgery patients in the United States (ClinicalTrials.gov identifier NCT00991341).

Effect of leukoreduction. Seventy percent of transfused RBCs in the United States in 2006 were leukocyte reduced (52). The potential benefits of using leukocyte-reduced blood include decreased transmission of viruses, febrile nonhemolytic transfusion reactions, HLA alloimmunization and platelet refractoriness, RBC alloimmunization, nosocomial infections, and death. Many of these effects may be mediated by transfusion-related immunomodulation (TRIM), a term describing immune activation or tolerance induction after blood transfusion, postulated to be due to infusion of donor leukocytes and released bioactive soluble factors. After implementation of universal leukoreduction in Canada, Hébert and colleagues reported decreased in-hospital mortality (odds ratio, 0.87) (72). However, a subsequent meta-analysis of before-and-after studies did not show any effect of leukoreduction on postoperative infection or mortality, after adjusting for confounding factors (73). In trauma patients, before-and-after cohort studies have shown decreased rates of nosocomial infection and ARDS (74, 75), but randomized clinical trials have shown no effect of leukoreduction on these outcomes (76, 77).

Transfusion thresholds. Only one randomized controlled trial in the general adult ICU population addresses appropriate RBC transfusion thresholds. In the Transfusion Requirements in Critical Care (TRICC) trial (49), 838 euvolemic patients without chronic anemia, myocardial ischemia, or on-going bleeding were randomized to either a restrictive or liberal transfusion strategy (threshold hemoglobin, 7 vs. 10 g/dl). No difference in the primary outcome of all-cause 30-day mortality was observed between treatment arms. Subgroup analyses identified patients less than 55 years old and with APACHE II scores less than 20 as having decreased 30-day mortality with a restrictive strategy. Although results of this trial have affected both guidelines and common practice, controversy still exists regarding specific patient groups: the elderly, and those with cardiovascular disease, with difficulty being liberated from mechanical ventilation, and in the early phase of septic shock.

In the 257 patients in the TRICC trial with ischemic heart disease, mortality was higher in the restrictive group (26 vs. 21%), although the difference was not statistically significant (78). A randomized controlled noninferiority trial of a liberal transfusion strategy (goal, hematocrit $\geq$ 30%) versus a restrictive strategy (goal, hematocrit $\geq$ 24%) after cardiac surgery showed no difference in 30-day all-cause mortality (79). Similarly, a randomized trial of RBC transfusion for Hb less than 10 g/dl or Hb less than 8 g/dl in 2,016 patients with cardiovascular disease or risk factors after hip fracture surgery (mean patient age, 82 yr) found no difference in mortality or ability to walk independently at 60 days follow-up (80). Several groups have examined the association of transfusions and mortality in large data sets of patients with acute coronary syndromes. Of these, one found transfusions to have a beneficial effect on survival when the hematocrit was less than 33% (81). In contrast, four studies found transfusions to be an independent predictor of greater short-term mortality (82–85); one identified a threshold hematocrit of 25%, above which transfusions were associated with increased risk of death (84). It is difficult to exclude confounding in such studies, and further trials of transfusion thresholds among patients with ischemic heart disease are needed.

As mentioned earlier, anemia is independently associated with extubation failure (42). In 10 anemic patients with stable severe COPD, Schönhofer and colleagues found that transfusion to a goal hemoglobin greater than 11 g/dl decreased ventilation and work of breathing (33). In a study of five anemic patients with COPD (mean hemoglobin, 8.7 g/dl) who were unable to be liberated from mechanical ventilation (28-d mean duration of ventilation; range, 13 to 49 d) (86), all were successfully extubated within 4 days of being transfused to a mean hemoglobin level of 12.4 g/dl. The TRICC trial included 713 patients on mechanical ventilation, of whom 219 were ventilated for greater than 7 days. In these subgroups, there was no difference in duration of mechanical ventilation or mortality between the two transfusion strategies (87). This analysis had power only to detect 25% differences in duration of mechanical ventilation, so a clinically important difference may have been missed. Transfusion is most likely to be beneficial in patients with the most severe ventilatory impairment and respiratory muscle weakness, and it remains to be determined whether and when a more liberal transfusion strategy is warranted in these patients.

Rivers and colleagues randomized 263 patients with severe sepsis or septic shock to standard therapy or early goal-directed therapy (EGDT) and found that EGDT significantly decreased mortality (88). Their protocol calls for RBC transfusion with a goal hematocrit of 30% if central venous oxygen saturation remains less than 70% after reaching goal central venous pressure and mean arterial pressure. Although the protocol as a whole improves survival, it is unclear which components are most effective, especially given prior evidence that blood transfusions may not improve tissue oxygenation in septic patients. A secondary analysis of patients with sepsis, shock, and ALI enrolled in the ARDSNet Fluid and Catheter Treatment Trial (89) found no difference in mortality based on transfusion status, and physiological criteria derived from the Rivers trial did not identify patients more likely to benefit from transfusion. However, study power was limited, with a minimal detectable mortality difference of 19%. A cohort study of 160 patients with septic shock found both transfusions and delayed EGDT to be risk factors for development of acute lung injury (90). A multicenter randomized trial called Protocolized Care for Early Septic Shock (ProCESS) is underway and may shed light on the role of RBC transfusion in the resuscitation of patients with septic shock. (ClinicalTrials.gov identifier NCT00510835).

Clinical transfusion guidelines. On the basis of available data, we would conclude that in most critically ill patients, a "restrictive" strategy of RBC transfusion (transfusion at Hb $<$ 7 g/dl) is preferable to a "liberal" transfusion strategy (transfusion at Hb $<$ 10 g/dl). In the absence of level 1 evidence, clinicians may consider RBC transfusion at higher Hb levels in certain clinical situations (*see* Table 1). Evidence-based guidelines regarding the use of RBC transfusion in critically ill adults developed by a joint taskforce of the Society of Critical Care Medicine (SCCM) and the Eastern Association for the Surgery of Trauma (EAST) discourage use of hemoglobin level as a "trigger" for transfusion, and recommend basing the decision on an individual patient's clinical condition and cardiopulmonary physiological parameters (91).

Erythropoietin

The finding that critically ill patients have a multifactorial blunted EPO response to anemia, as described above, led to interest in whether treatment with EPO could improve outcomes. Several trials have addressed this question; but three by Corwin and colleagues comprise the vast majority of patients enrolled. The first (EPO-1) was a pilot study of 160 adults in a multidisciplinary ICU (92). Exclusion criteria were extensive, and included vasopressor requirement and high levels of ventilatory support. The intervention group received EPO at 300 units/kg daily for 5 days, followed by every other day dosing (mean dose, 138,000

TABLE 1. SCENARIOS IN WHICH A MORE LIBERAL RBC TRANSFUSION STRATEGY MAY BE REASONABLE

Clinical Situation	Cardiopulmonary Parameters
Active myocardial ischemia	Tachycardia, elevated cardiac index
Difficulty being liberated from mechanical ventilation	Severe ventilatory impairment, respiratory muscle weakness, high minute ventilation
Early phase of septic shock	Central venous pressure 8–12 mm Hg, mean arterial pressure $\geq$ 65 mm Hg, *and* central venous oxygen saturation $<$ 70%

In most critically ill patients, a "restrictive" strategy of RBC transfusion (transfusion at Hb $<$ 7 g/dl) is preferable to a "liberal" transfusion strategy (transfusion at Hb $<$ 10 g/dl). In the absence of level 1 evidence, based on physiologic rationale and available data, clinicians may consider RBC transfusion at Hb $>$ 7 g/dl in certain clinical situations, especially in the presence of certain cardiopulmonary parameters, described above.

units in the first week of therapy). The second study (EPO-2) was more inclusive in terms of entry criteria; it included 1,302 patients in 65 U.S. medical centers and used a lower dose of 40,000 units weekly (93). Neither trial used a transfusion threshold protocol; the mean pretransfusion hematocrit was 27% (Hb, ~9 g/dl) in the first study and a Hb level of 8.5 g/dl in the second. In both studies, the intervention group received significantly fewer RBC transfusions (20–30% in the first 28 d) with maintenance of a higher hemoglobin concentration, but no other clinical benefit or harm was identified. The third trial (EPO-3) enrolled 1,460 patients, and also used a dose of 40,000 units weekly (50). In contrast, there was no difference seen between rates of RBC transfusion in the two groups. This may be related to a more restrictive transfusion practice (mean pretransfusion hemoglobin, 8.0 g/dl). Moreover, the intervention group had a higher rate of thrombotic events (hazard ratio, 1.41), although in post-hoc analysis this risk was not increased among patients receiving standard prophylactic or therapeutic doses of heparin.

Although overall mortality between the two groups in EPO-3 was not different, subgroup multivariate Cox regression analysis of the 793 trauma patients showed lower adjusted mortality in the EPO group on Day 29 (adjusted hazard ratio, 0.37; 95% confidence interval, 0.19 to 0.72). However, as the interaction between the stratification variables of the admission group and the study group was not significant, the significance of this finding has been questioned (94). Although clinical trials have not clearly shown benefit with EPO, animal models of hemorrhagic shock and organ ischemia–reperfusion injuries demonstrate cytoprotective effects of EPO independent of its hematopoietic effects. These studies used much higher doses of EPO than in human trials. This may be necessary for organ protection, because the receptor that mediates the cytoprotective effects of EPO (EPO-BCR) has a lower affinity for EPO than the receptor that mediates hematopoiesis (EPO-R). Novel peptides derived from EPO that retain its cytoprotective properties but lack its hematopoietic and prothrombotic effects have been developed and are currently under investigation (95).

The majority of data indicate that in the current atmosphere of restrictive transfusion practice for critically ill patients, erythropoietin, in the form (epoetin alfa) and at the dose and frequency used in the 2007 study by Corwin and colleagues (50), does not improve survival, and may increase risk of thrombotic complications in those not given prophylaxis for deep venous thrombosis. Although the findings surrounding EPO use in critically ill trauma patients are intriguing, carefully designed clinical trials are required before its use in this population can be justified. Given the known hazards of RBC transfusion and ineffectiveness of blood substitutes (*see below*), there remains a need to explore alternative erythropoiesis-stimulating agents. Such possibilities include agents that increase concentrations of endogenous HIF, a transcription factor that regulates several genes involved in erythropoiesis, including EPO and its receptor (32).

Blood Substitutes

The impetus to develop "blood substitutes" includes concerns about blood shortages, which are expected to become more problematic with changing population demographics, and the various shortcomings of stored blood. Two types of oxygen carriers have been developed: cell-free hemoglobin-based oxygen carriers (HBOCs) and perfluorocarbons. Early preparations of HBOCs caused nephrotoxicity, and even in the most recent generation of products, there is serious concern about vasoactivity (from avid NO scavenging by nonencapsulated hemoglobin), impaired perfusion, and increased rates of myocardial infarction and death (96–98). No "blood substitutes" are currently approved for human use in the United States. Of the two commercially available HBOCs, Oxyglobin is licensed for veterinary use in the United States and Hemopure is licensed for human use in South Africa. The only available perfluorocarbon is Perftoran, which is approved for human use in Russia and Mexico. Studies of artificial oxygen carriers have targeted primarily patients suffering acute hemorrhage from surgery or trauma. Even aside from their potential adverse effects, these agents, as currently formulated, would be of limited use for anemia related to the inflammation of critical illness because of their short intravascular half-life of 12 to 48 hours.

Iron Therapy

Iron has been shown to promote the growth and virulence of a number of microbes responsible for nosocomial infections (99). It is theorized that low serum iron in acute inflammation is a protective host response to impair bacterial growth. As a result, concern exists for greater infection rates with iron supplementation. Although this outcome is biologically plausible and grounded in animal studies, there is scant evidence in human studies (99). The issue has been examined most extensively in chronic hemodialysis patients (100) and multiple studies have failed to show any increased risk of infection associated with iron therapy.

Few studies have examined iron supplementation in the critical care population. One retrospective analysis in surgery patients identified 27 who received intravenous iron therapy, and found that compared with matched control subjects, these patients did not have higher rates of bacteremia (101). In 863 patients postcardiopulmonary bypass surgery, treated with both intravenous iron and erythropoietin as needed, or with blood transfusions, there was no difference in subsequent infection rate (102). In a trial of 200 patients receiving care in a surgical ICU (103), randomization to enteral ferrous sulfate (vs. placebo) failed to produce any statistically significant difference in hematocrit, iron markers, infection rates, antibiotic days, hospital length of stay, or mortality. Notably, patients given iron were significantly less likely to receive a blood transfusion (29.9 vs. 44.7%; $P = 0.03$) as compared with the placebo group. On

subgroup analysis, this effect was restricted to patients with baseline iron-deficient erythropoiesis as defined by elevated zinc protoporphyrin concentration. There is an ongoing multicenter randomized clinical trial of intravenous iron for the treatment of anemia in critically ill trauma patients (ClinicalTrials.gov identifier NCT01180894).

Intravenous iron supplementation may have better efficacy than enteral administration because of the block of intestinal absorption by hepcidin. It has been shown that iron may be useful in heart failure and pulmonary hypertension (104, 105), independent of changes in hematocrit. Further research, however, is needed before iron supplementation can be recommended for the routine care of anemic critically ill patients, owing to the potentially heightened infection risk.

Minimization of Blood Loss

Strategies to minimize blood loss include the use of small-volume phlebotomy tubes, point-of-care testing and noninvasive testing, the reinfusion of discard sample from indwelling lines, and the elimination of unnecessary laboratory studies.

"Small-volume" phlebotomy tubes typically require less than 2 ml of blood, and sometimes as little as 0.5 ml. The use of small-volume or pediatric tubes for ICU patients, either as a single intervention (106, 107) or in combination with other blood conservation measures (108, 109), can reduce blood loss by this route by 33 to 80%. Point-of-care blood analysis provides test results with minimal delay and often requires samples of less than 0.5 ml. Noninvasive monitoring, such as pulse oximetry and end-tidal CO_2 monitoring in select patients, or oximeters capable of measuring hemoglobin noninvasively (110), may further reduce blood loss due to phlebotomy.

The presence of an indwelling arterial catheter is associated with a 33% increase in number of blood tests performed and a 44% increase in amount of blood removed from the patient, even after accounting for severity of illness (111). This may be due to the perceived ease of phlebotomy and the need to discard 2 to 10 ml of "blood" to clear the catheter of infusate. Such waste can be eliminated by returning this blood to the patient, either via three-way stopcock or a commercially available blood-sampling system. Using such a closed system decreases blood loss (109, 112) and one before-and-after study also showed reduced amount of blood transfusions (0.07 vs. 0.13 units/patient/day; $P = 0.02$), and even reduced hospital mortality (30 vs. 53%; $P = 0.001$) (113).

It is possible to reduce the number of laboratory studies in ICU patients without compromising quality of care (114–116). This training has been termed "learning to not know" (116). Such educational initiatives should be cooperative multidisciplinary projects, with ongoing training and feedback to promote durability. Clinicians should also be encouraged to group laboratory tests to minimize the number of phlebotomies. Surveys of Australian and European ICUs (109, 117) show that only a small minority of institutions routinely use the blood conservation techniques described above; thus an important opportunity exists to potentially improve patient outcomes by minimizing phlebotomy.

CONCLUSIONS

A growing body of literature on anemia of critical illness points to four conclusions: (*1*) anemia is highly prevalent in the critically ill; (*2*) it is associated with higher health care resource use; (*3*) it may be associated with poor patient outcomes; and (*4*) there is no currently available therapy without shortcomings. Further research is needed to delineate risks, benefits, and effectiveness of various management strategies in specific patient populations. While awaiting further evidence, intensivists should pay careful attention to minimizing blood loss whenever possible, and tailoring the management of anemia to the needs of each patient.

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References

1. Vincent JL, Baron JF, Reinhart K, Gattinoni L, Thijs L, Webb A, Meier-Hellmann A, Nollet G, Peres-Bota D; ABC (Anemia and Blood Transfusion in Critical Care) Investigators. Anemia and blood transfusion in critically ill patients. *JAMA* 2002;288:1499–1507.
2. Corwin HL, Gettinger A, Pearl RG, Fink MP, Levy MM, Abraham E, MacIntyre NR, Shabot MM, Duh MS, Shapiro MJ. The CRIT Study: anemia and blood transfusion in the critically ill—current clinical practice in the United States. *Crit Care Med* 2004;32:39–52.
3. Thomas J, Jensen L, Nahirniak S, Gibney RT. Anemia and blood transfusion practices in the critically ill: a prospective cohort review. *Heart Lung* 2010;39:217–225.
4. Baskurt OK, Yalcin O, Meiselman HJ. Hemorheology and vascular control mechanisms. *Clin Hemorheol Microcirc* 2004;30:169–178.
5. Cosby K, Partovi KS, Crawford JH, Patel RP, Reiter CD, Martyr S, Yang BK, Waclawiw MA, Zalos G, Xu X, *et al*. Nitrite reduction to nitric oxide by deoxyhemoglobin vasodilates the human circulation. *Nat Med* 2003;9:1498–1505.
6. Hess DT, Matsumoto A, Kim SO, Marshall HE, Stamler JS. Protein S-nitrosylation: purview and parameters. *Nat Rev Mol Cell Biol* 2005;6:150–166.
7. Sprague RS, Ellsworth ML, Stephenson AH, Lonigro AJ. ATP: the red blood cell link to NO and local control of the pulmonary circulation. *Am J Physiol* 1996;271:H2717–H2722.
8. Kurata M, Suzuki M, Agar NS. Antioxidant systems and erythrocyte life-span in mammals. *Comp Biochem Physiol B* 1993;106:477–487.
9. Hillman RS, Henderson PA. Control of marrow production by the level of iron supply. *J Clin Invest* 1969;48:454–460.
10. Stäubli M, Ott P, Waber U, Stäuble UP, Jeanneret C, Peheim E, Straub PW. Erythrocyte adenosine triphosphate depletion during voluntary hyperventilation. *J Appl Physiol* 1985;59:1196–1200.
11. Sakr Y, Dubois MJ, De Backer D, Creteur J, Vincent JL. Persistent microcirculatory alterations are associated with organ failure and death in patients with septic shock. *Crit Care Med* 2004;32:1825–1831.
12. Myssina S, Huber SM, Birka C, Lang PA, Lang KS, Friedrich B, Risler T, Wieder T, Lang F. Inhibition of erythrocyte cation channels by erythropoietin. *J Am Soc Nephrol* 2003;14:2750–2757.
13. Lang F, Lang KS, Lang PA, Huber SM, Wieder T. Mechanisms and significance of eryptosis. *Antioxid Redox Signal* 2006;8:1183–1192.
14. Rice L, Alfrey CP. The negative regulation of red cell mass by neocytolysis: physiologic and pathophysiologic manifestations. *Cell Physiol Biochem* 2005;15:245–250.
15. Corwin HL, Parsonnet KC, Gettinger A. RBC transfusion in the ICU: is there a reason? *Chest* 1995;108:767–771.
16. Barie PS. Phlebotomy in the intensive care unit: strategies for blood conservation. *Crit Care* 2004;8:S34–S36.
17. Hadfield RJ, Sinclair DG, Houldsworth PE, Evans TW. Effects of enteral and parenteral nutrition on gut mucosal permeability in the critically ill. *Am J Respir Crit Care Med* 1995;152:1545–1548.
18. Cook D, Heyland D, Griffith L, Cook R, Marshall J, Pagliarello J; Canadian Critical Care Trials Group. Risk factors for clinically important upper gastrointestinal bleeding in patients requiring mechanical ventilation. *Crit Care Med* 1999;27:2812–2817.
19. Rodriguez RM, Corwin HL, Gettinger A, Corwin MJ, Gubler D, Pearl RG. Nutritional deficiencies and blunted erythropoietin response as causes of the anemia of critical illness. *J Crit Care* 2001;16:36–41.
20. Fleming RE, Bacon BR. Orchestration of iron homeostasis. *N Engl J Med* 2005;352:1741–1744.
21. Weiss G, Goodnough LT. Anemia of chronic disease. *N Engl J Med* 2005;352:1011–1023.

22. Nemeth E, Rivera S, Gabayan V, Keller C, Taudorf S, Pedersen BK, Ganz T. IL-6 mediates hypoferremia of inflammation by inducing the synthesis of the iron regulatory hormone hepcidin. *J Clin Invest* 2004;113:1271–1276.
23. Corwin HL, Krantz SB. Anemia of the critically ill: "acute" anemia of chronic disease. *Crit Care Med* 2000;28:3098–3099.
24. Fonseca RB, Mohr AM, Wang L, Sifri ZC, Rameshwar P, Livingston DH. The impact of a hypercatecholamine state on erythropoiesis following severe injury and the role of IL-6. *J Trauma* 2005;59:884–889.
25. von Ahsen N, Müller C, Serke S, Frei U, Eckardt KU. Important role of nondiagnostic blood loss and blunted erythropoietic response in the anemia of medical intensive care patients. *Crit Care Med* 1999;27: 2630–2639.
26. Rogiers P, Zhang H, Leeman M, Nagler J, Neels H, Mélot C, Vincent JL. Erythropoietin response is blunted in critically ill patients. *Intensive Care Med* 1997;23:159–162.
27. Krafte-Jacobs B. Anemia of critical illness and erythropoietin deficiency. *Intensive Care Med* 1997;23:137–138.
28. Bakris GL, Sauter ER, Hussey JL, Fisher JW, Gaber AO, Winsett R. Effects of theophylline on erythropoietin production in normal subjects and in patients with erythrocytosis after renal transplantation. *N Engl J Med* 1990;323:86–90.
29. Linde T, Sandhagen B, Hägg A, Mörlin C, Danielson BG. Decreased blood viscosity and serum levels of erythropoietin after antihypertensive treatment with amlodipine or metoprolol: results of a cross-over study. *J Hum Hypertens* 1996;10:199–205.
30. Vlahakos DV, Marathias KP, Madias NE. The role of the renin–angiotensin system in the regulation of erythropoiesis. *Am J Kidney Dis* 2010;56:558–565.
31. Weiskopf RB, Viele MK, Feiner J, Kelley S, Lieberman J, Noorani M, Leung JM, Fisher DM, Murray WR, Toy P, *et al*. Human cardiovascular and metabolic response to acute, severe isovolemic anemia. *JAMA* 1998;279:217–221.
32. Semenza GL. Regulation of oxygen homeostasis by hypoxia-inducible factor 1. *Physiology (Bethesda)* 2009;24:97–106.
33. Schönhofer B, Wenzel M, Geibel M, Köhler D. Blood transfusion and lung function in chronically anemic patients with severe chronic obstructive pulmonary disease. *Crit Care Med* 1998;26:1824–1828.
34. Winslow RM, Monge CC, Brown EG, Klein HG, Sarnquist F, Winslow NJ, McKneally SS. Effects of hemodilution on O_2 transport in high-altitude polycythemia. *J Appl Physiol* 1985;59:1495–1502.
35. Culleton BF, Manns BJ, Zhang J, Tonelli M, Klarenbach S, Hemmelgarn BR. Impact of anemia on hospitalization and mortality in older adults. *Blood* 2006;107:3841–3846.
36. Martinez FJ, Foster G, Curtis JL, Criner G, Weinmann G, Fishman A, DeCamp MM, Benditt J, Sciurba F, Make B, *et al*.; NETT Research Group. Predictors of mortality in patients with emphysema and severe airflow obstruction. *Am J Respir Crit Care Med* 2006;173:1326–1334.
37. Chambellan A, Chailleux T, Similowski T. Prognostic value of the hematocrit in patients with severe COPD receiving long term oxygen therapy. *Chest* 2005;128:1201–1208.
38. Similowski T, Agustí A, MacNee W, Schönhofer B. The potential impact of anaemia of chronic disease in COPD. *Eur Respir J* 2006;27: 390–396.
39. Go AS, Yang J, Ackerson LM, Lepper K, Robbins S, Massie BM, Shlipak MG. Hemoglobin level, chronic kidney disease, and the risks of death and hospitalization in adults with chronic heart failure: the Anemia in Chronic Heart Failure: Outcomes and Resource Utilization (ANCHOR) Study. *Circulation* 2006;113:2713–2723.
40. Salisbury AC, Alexander KP, Reid KJ, Masoudi FA, Rathore SS, Wang TY, Bach RG, Marso SP, Spertus JA, Kosiborod M. Incidence, correlates, and outcomes of acute, hospital-acquired anemia in patients with acute myocardial infarction. *Circ Cardiovasc Qual Outcomes* 2010;3:337–346.
41. Mehdi U, Toto R. Anemia, diabetes, and chronic kidney disease. *Diabetes Care* 2009;32:1320–1326.
42. Khamiees M, Raju P, DeGirolamo A, Amoateng-Adjepong Y, Manthous CA. Predictors of extubation outcome in patients who have successfully completed a spontaneous breathing trial. *Chest* 2001;120:1262–1270.
43. Thygesen K, Alpert JS, White HD; Joint ESC/ACCF/AHA/WHF Task Force for the Redefinition of Myocardial Infarction. Universal definition of myocardial infarction. *Eur Heart J* 2007;28:2525–2538.
44. Rasmussen L, Christensen S, Lenler-Petersen P, Johnsen SP. Anemia and 90-day mortality in COPD patients requiring invasive mechanical ventilation. *Clin Epidemiol.* 2010;3:1–5.
45. Carson JL, Noveck H, Berlin JA, Gould SA. Mortality and morbidity in patients with very low postoperative Hb levels who decline blood transfusion. *Transfusion* 2002;42:812–818.
46. Karon BS, Griesmann L, Scott R, Bryant SC, Dubois JA, Shirey TL, Presti S, Santrach PJ. Evaluation of the impact of hematocrit and other interference on the accuracy of hospital-based glucose meters. *Diabetes Technol Ther* 2008;10:111–120.
47. Weiskopf RB, Kramer JH, Viele M, Neumann M, Feiner JR, Watson JJ, Hopf HW, Toy P. Acute severe isovolemic anemia impairs cognitive function and memory in humans. *Anesthesiology* 2000;92: 1646–1652.
48. Bateman AP, McArdle F, Walsh TS. Time course of anemia during six months follow up following intensive care discharge and factors associated with impaired recovery of erythropoiesis. *Crit Care Med* 2009;37:1906–1912.
49. Hébert PC, Wells G, Blajchman MA, Marshall J, Martin C, Pagliarello G, Tweeddale M, Schweitzer I, Yetisir E; Transfusion Requirements in Critical Care Investigators, Canadian Critical Care Trials Group. A multicenter, randomized, controlled clinical trial of transfusion requirements in critical care. *N Engl J Med* 1999;340:409–417.
50. Corwin HL, Gettinger A, Fabian TC, May A, Pearl RG, Heard S, An R, Bowers PJ, Burton P, Klausner MA, *et al*.; EPO Critical Care Trials Group. Efficacy and safety of epoetin alfa in critically ill patients. *N Engl J Med* 2007;357:965–976.
51. Vincent JL, Sakr Y, Sprung C, Harboe S, Damas P; Sepsis Occurrence in Acutely Ill Patients (SOAP) Investigators. Are blood transfusions associated with greater mortality rates? *Anesthesiology* 2008;108:31–39.
52. Whitaker BI, Henry RA. The 2007 nationwide blood collection and utilization survey report [Internet]. Washington, DC: Department of Health and Human Services; c2008 [accessed 2011 Oct 26]. Available from: http://www.hhs.gov/ash/bloodsafety/2007nbcus_survey.pdf
53. Conrad SA, Dietrich KA, Hebert CA, Romero MD. Effect of red cell transfusion on oxygen consumption following fluid resuscitation in septic shock. *Circ Shock* 1990;31:419–429.
54. Marik PE, Sibbald WJ. Effect of stored-blood transfusion on oxygen delivery in patients with sepsis. *JAMA* 1993;269:3024–3029.
55. Ronco JJ, Phang PT, Walley KR, Wiggs B, Fenwick JC, Russell JA. Oxygen consumption is independent of changes in oxygen delivery in severe adult respiratory distress syndrome. *Am Rev Respir Dis* 1991;143:1267–1273.
56. Shah DM, Gottlieb ME, Rahm RL, Stratton HH, Barie PS, Paloski WH, Newell JC. Failure of red blood cell transfusion to increase oxygen transport or mixed venous Po_2 in injured patients. *J Trauma* 1982;22:741–746.
57. Gonzalez AM, Yazici I, Kusza K, Siemionow M. Effects of fresh versus banked blood transfusions on microcirculatory hemodynamics and tissue oxygenation in the rat cremaster model. *Surgery* 2007;141:630–639.
58. van Bommel J, de Korte D, Lind A, Siegemund M, Trouwborst A, Verhoeven AJ, Ince C, Henny CP. The effect of the transfusion of stored RBCs on intestinal microvascular oxygenation in the rat. *Transfusion* 2001;41:1515–1523.
59. Heaton A, Keegan T, Holme S. *In vivo* regeneration of red cell 2,3-diphosphoglycerte following transfusion of DPG-depleted AS-1, AS-3 and CPDA-1 red cells. *Br J Haematol* 1989;71:131–136.
60. Dodd RY. Current risk for transfusion transmitted infections. *Curr Opin Hematol* 2007;14:671–676.
61. Skeate RC, Eastlund T. Distinguishing between transfusion related acute lung injury and transfusion associated circulatory overload. *Curr Opin Hematol* 2007;14:682–687.
62. Vamvakas EC, Blajchman MA. Transfusion-related immunomodulation (TRIM): an update. *Blood Rev* 2007;21:327–348.
63. Marik PE, Corwin HL. Efficacy of red blood cell transfusion in the critically ill: a systematic review of the literature. *Crit Care Med* 2008;36:2667–2674.

64. Chaiwat O, Lang JD, Vavilala MS, Wang J, MacKenzie EJ, Jurkovich GJ, Rivara FP. Early packed red blood cell transfusion and acute respiratory distress syndrome after trauma. *Anesthesiology* 2009;110: 351–360.
65. Reynolds JD, Ahearn GS, Angelo M, Zhang J, Cobb F, Stamler JS. *S*-Nitrosohemoglobin deficiency: a mechanism for loss of physiological activity in banked blood. *Proc Natl Acad Sci USA* 2007;104: 17058–17062.
66. Donadee C, Raat NJ, Kanias T, Tejero J, Lee JS, Kelley EE, Zhao X, Liu C, Reynolds H, Azarov I, *et al*. Nitric oxide scavenging by red blood cell microparticles and cell-free hemoglobin as a mechanism for the red cell storage lesion. *Circulation* 2011;124:465–476.
67. Xiong Z, Cavaretta J, Qu L, Stolz DB, Triulzi D, Lee JS. Red blood cell microparticles show altered inflammatory chemokine binding and release ligand upon interaction with platelets. *Transfusion* 2011;51: 610–621.
68. Lelubre C, Piagnerelli M, Vincent JL. Association between duration of storage of transfused red blood cells and morbidity and mortality in adult patients: myth or reality? *Transfusion* 2009;49:1384–1394.
69. Edgren G, Kamper-Jørgensen M, Eloranta S, Rostgaard K, Custer B, Ullum H, Murphy EL, Busch MP, Reilly M, Melbye M, *et al*. Duration of red blood cell storage and survival of transfused patients. *Transfusion* 2010;50:1185–1195.
70. Hod EA, Zhang N, Sokol SA, Wojczyk BS, Francis RO, Ansaldi D, Francis KP, Della-Latta P, Whittier S, Sheth S, *et al*. Transfusion of red blood cells after prolonged storage produces harmful effects that are mediated by iron and inflammation. *Blood* 2010;115:4284–4292.
71. Vlaar AP, Hofstra JJ, Levi M, Kulik W, Nieuwland R, Tool AT, Schultz MJ, de Korte D, Juffermans NP. Supernatant of aged erythrocytes causes lung inflammation and coagulopathy in a "two-hit" *in vivo* syngeneic transfusion model. *Anesthesiology* 2010;113: 92–103.
72. Hébert PC, Fergusson D, Blajchman MA, Wells GA, Kmetic A, Coyle D, Heddle N, Germain M, Goldman M, Toye B, *et al*.; Leukoreduction Study Investigators. Clinical outcomes following institution of the Canadian universal leukoreduction program for red blood cell transfusions. *JAMA* 2003;289:1941–1949.
73. Vamvakas EC. White-blood-cell-containing allogeneic blood transfusion, postoperative infection and mortality: a meta-analysis of observational "before-and-after" studies. *Vox Sang* 2004;86:111–119.
74. Friese RS, Sperry JL, Phelan HA, Gentilello LM. The use of leukoreduced red blood cell products is associated with fewer infectious complications in trauma patients. *Am J Surg* 2008;196:56–61.
75. Plurad D, Belzberg H, Schulman I, Green D, Salim A, Inaba K, Rhee P, Demetriades D. Leukoreduction is associated with a decreased incidence of late onset acute respiratory distress syndrome after injury. *Am Surg* 2008;74:117–123.
76. Watkins TR, Rubenfeld GD, Martin TR, Nester TA, Caldwell E, Billgren J, Ruzinski J, Nathens AB. Effects of leukoreduced blood on acute lung injury after trauma: a randomized controlled trial. *Crit Care Med* 2008;36:1493–1499.
77. Nathens AB, Nester TA, Rubenfeld GD, Nirula R, Gernsheimer TB. The effects of leukoreduced blood transfusion on infection risk following injury: a randomized controlled trial. *Shock* 2006;26:342–347.
78. Hébert PC, Yetisir E, Martin C, Blajchman MA, Wells G, Marshall J, Tweeddale M, Pagliarello G, Schweitzer I; Transfusion Requirements in Critical Care Investigators for the Canadian Critical Care Trials Group. Is a low transfusion threshold safe in critically ill patients with cardiovascular diseases? *Crit Care Med* 2001;29:227–234.
79. Hajjar LA, Vincent JL, Galas FR, Nakamura RE, Silva CM, Santos MH, Fukushima J, Kalil Filho R, Sierra DB, Lopes NH, *et al*. Transfusion requirements after cardiac surgery: the TRACS randomized controlled trial. *JAMA* 2010;304:1559–1567.
80. Carson JL, Terrin ML, Noveck H, Sanders DW, Chaitman BR, Rhoads GG, Nemo G, Dragert K, Beaupre L, Hildebrand K, Macaulay W, Lewis C, Cook DR, Dobbin G, Zakriya KJ, Apple FS, Horney RA, Magaziner J; FOCUS Investigators. Liberal or restrictive transfusion in high-risk patients after hip surgery. *N Engl J Med* 2011;365:2453–2462.
81. Wu WC, Rathore SS, Wang Y, Radford MJ, Krumholz HM. Blood transfusion in elderly patients with acute myocardial infarction. *N Engl J Med* 2001;345:1230–1236.
82. Jolicoeur EM, O'Neill WW, Hellkamp A, Hamm CW, Holmes DR Jr, Al-Khalidi HR, Patel MR, Van de Werf FJ, Pieper K, Armstrong PW, *et al*. APEX-AMI Investigators. Transfusion and mortality in patients with ST-segment elevation myocardial infarction treated with primary percutaneous coronary intervention. *Eur Heart J* 2009; 30:2575–2583.
83. Nikolsky E, Mehran R, Sadeghi HM, Grines CL, Cox DA, Garcia E, Tcheng JE, Griffin JJ, Guagliumi G, Stuckey T, *et al*. Prognostic impact of blood transfusion after primary angioplasty for acute myocardial infarction: analysis from the CADILLAC (Controlled Abciximab and Device Investigation to Lower Late Angioplasty Complications) Trial. *JACC Cardiovasc Interv* 2009;2:624–632.
84. Rao SV, Jollis JG, Harrington RA, Granger CB, Newby LK, Armstrong PW, Moliterno DJ, Lindblad L, Pieper K, Topol EJ, *et al*. Relationship of blood transfusion and clinical outcomes in patients with acute coronary syndromes. *JAMA* 2004;292:1555–1562.
85. Yang X, Alexander KP, Chen AY, Roe MT, Brindis RG, Rao SV, Gibler WB, Ohman EM, Peterson ED; CRUSADE Investigators. The implications of blood transfusions for patients with non–ST-segment elevation acute coronary syndromes: results from the CRUSADE National Quality Improvement Initiative. *J Am Coll Cardiol* 2005;46:1490–1495.
86. Schönhofer B, Böhrer H, Köhler D. Blood transfusion facilitating difficult weaning from the ventilator. *Anaesthesia* 1998;53:181–184.
87. Hébert PC, Blajchman MA, Cook DJ, Yetisir E, Wells G, Marshall J, Schweitzer I; Transfusion Requirements in Critical Care Investigators for the Canadian Critical Care Trials Group. Do blood transfusions improve outcomes related to mechanical ventilation? *Chest* 2001;119:1850–1857.
88. Rivers E, Nguyen B, Havstad S, Ressler J, Muzzin A, Knoblich B, Peterson E, Tomlanovich M; Early Goal-directed Therapy Collaborative Group. Early goal-directed therapy in the treatment of severe sepsis and septic shock. *N Engl J Med* 2001;345:1368–1377.
89. Parsons EC, Hough CL, Seymour CW, Cooke CR, Rubenfeld GD, Watkins TR, Network NA. Red blood cell transfusion and outcomes in patients with acute lung injury, sepsis and shock. *Crit Care* 2011; 15:R221.
90. Iscimen R, Cartin-Ceba R, Yilmaz M, Khan H, Hubmayr RD, Afessa B, Gajic O. Risk factors for the development of acute lung injury in patients with septic shock: an observational cohort study. *Crit Care Med* 2008;36:1518–1522.
91. Napolitano LM, Kurek S, Luchette FA, Corwin HL, Barie PS, Tisherman SA, Hebert PC, Anderson GL, Bard MR, Bromberg W, *et al*.; American College of Critical Care Medicine of the Society of Critical Care Medicine; Eastern Association for the Surgery of Trauma Practice Management Workgroup. Clinical practice guideline: red blood cell transfusion in adult trauma and critical care. *Crit Care Med* 2009;37:3124–3157.
92. Corwin HL, Gettinger A, Rodriguez RM, Pearl RG, Gubler KD, Enny C, Colton T, Corwin MJ. Efficacy of recombinant human erythropoietin in the critically ill patient: a randomized, double-blind, placebo-controlled trial. *Crit Care Med* 1999;27:2346–2350.
93. Corwin HL, Gettinger A, Pearl RG, Fink MP, Levy MM, Shapiro MJ, Corwin MJ, Colton T; EPO Critical Care Trials Group. Efficacy of recombinant human erythropoietin in critically ill patients: a randomized controlled trial. *JAMA* 2002;288:2827–2835.
94. Cook D, Crowther M. Targeting anemia with erythropoietin during critical illness. *N Engl J Med* 2007;357:1037–1039.
95. Solling C. Organ-protective and immunomodulatory effects of erythropoietin—an update on recent clinical trials. *Basic Clin Pharmacol Toxicol* 2012;110:113-121.
96. Moore EE, Moore FA, Fabian TC, Bernard AC, Fulda GJ, Hoyt DB, Duane TM, Weireter LJ Jr, Gomez GA, Cipolle MD, *et al*.; PolyHeme Study Group. Human polymerized hemoglobin for the treatment of hemorrhagic shock when blood is unavailable: the USA multicenter trial. *J Am Coll Surg* 2009;208:1–13.
97. Jahr JS, Mackenzie C, Pearce LB, Pitman A, Greenburg AG. HBOC-201 as an alternative to blood transfusion: efficacy and safety evaluation in a multicenter phase III trial in elective orthopedic surgery. *J Trauma* 2008;64:1484–1497.

98. Natanson C, Kern SJ, Lurie P, Banks SM, Wolfe SM. Cell-free hemoglobin-based blood substitutes and risk of myocardial infarction and death: a meta-analysis. *JAMA* 2008;299:2304–2312.
99. Weinberg ED. Iron loading and disease surveillance. *Emerg Infect Dis* 1999;5:346–352.
100. Hayat A. Safety issues with intravenous iron products in the management of anemia in chronic kidney disease. *Clin Med Res* 2008;6:93–102.
101. Swoboda SM, Lipsett PA. Intravenous iron as a risk factor for bacteremia in the surgical intensive care unit patient. *Surg Infect (Larchmt)* 2005;6:158.
102. Torres S, Kuo YH, Morris K, Neibart R, Holtz JB, Davis JM. Intravenous iron following cardiac surgery does not increase the infection rate. *Surg Infect (Larchmt)* 2006;7:361–366.
103. Pieracci FM, Henderson P, Rodney JR, Holena DN, Genisca A, Ip I, Benkert S, Hydo LJ, Eachempati SR, Shou J, *et al*. Randomized, double-blind, placebo-controlled trial of effects of enteral iron supplementation on anemia and risk of infection during surgical critical illness. *Surg Infect (Larchmt)* 2009;10:9–19.
104. Anker SD, Comin Colet J, Filippatos G, Willenheimer R, Dickstein K, Drexler H, Lüscher TF, Bart B, Banasiak W, Niegowska J, *et al*.; FAIR-HF Trial Investigators. Ferric carboxymaltose in patients with heart failure and iron deficiency. *N Engl J Med* 2009;361:2436–2448.
105. Smith TG, Talbot NP, Privat C, Rivera-Ch M, Nickol AH, Ratcliffe PJ, Dorrington KL, León-Velarde F, Robbins PA. Effects of iron supplementation and depletion on hypoxic pulmonary hypertension: two randomized controlled trials. *JAMA* 2009;302:1444–1450.
106. Smoller BR, Kruskall MS, Horowitz GL. Reducing adult phlebotomy blood loss with the use of pediatric-sized blood collection tubes. *Am J Clin Pathol* 1989;91:701–703.
107. Sanchez-Giron F, Alvarez-Mora F. Reduction of blood loss from laboratory testing in hospitalized adult patients using small-volume (pediatric) tubes. *Arch Pathol Lab Med* 2008;132:1916–1919.
108. Foulke GE, Harlow DJ. Effective measures for reducing blood loss from diagnostic laboratory tests in intensive care unit patients. *Crit Care Med* 1989;17:1143–1145.
109. Harber CR, Sosnowski KJ, Hegde RM. Highly conservative phlebotomy in adult intensive care—a prospective randomized controlled trial. *Anaesth Intensive Care* 2006;34:434–437.
110. Macknet MR, Allard M, Applegate RL II, Rook J. The accuracy of noninvasive and continuous total hemoglobin measurement by pulse CO-oximetry in human subjects undergoing hemodilution. *Anesth Analg* 2010;111:1424–1426.
111. Low LL, Harrington GR, Stoltzfus DP. The effect of arterial lines on blood-drawing practices and costs in intensive care units. *Chest* 1995;108:216–219.
112. Peruzzi WT, Parker MA, Lichtenthal PR, Cochran-Zull C, Toth B, Blake M. A clinical evaluation of a blood conservation device in medical intensive care unit patients. *Crit Care Med* 1993;21:501–506.
113. Mukhopadhyay A, Yip HS, Prabhuswamy D, Chan YH, Phua J, Lim TK, Leong P. The use of a blood conservation device to reduce red blood cell transfusion requirements: a before and after study. *Crit Care* 2010;14:R7.
114. Prat G, Lefèvre M, Nowak E, Tonnelier JM, Renault A, L'Her E, Boles JM. Impact of clinical guidelines to improve appropriateness of laboratory tests and chest radiographs. *Intensive Care Med* 2009;35:1047–1053.
115. Roberts DE, Bell DD, Ostryzniuk T, Dobson K, Oppenheimer L, Martens D, Honcharik N, Cramp H, Loewen E, Bodnar S, *et al*. Eliminating needless testing in intensive care—an information-based team management approach. *Crit Care Med* 1993;21:1452–1458.
116. Barie PS, Hydo LJ. Lessons learned: durability and progress of a program for ancillary cost reduction in surgical critical care. *J Trauma* 1997;43:590–594.
117. O'Hare D, Chilvers RJ. Arterial blood sampling practices in intensive care units in England and Wales. *Anaesthesia* 2001;56:568–571.