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Supra-plasma expanders – the future of treating blood loss and anemia without red cell transfusions?

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Anemia resulting from clinical conditions or loss of blood is treated by default with blood transfusion. However, securing the required blood and ensuring its availability and reserves adequate for the practice of modern medicine is a daunting task, one whose costs and risks of deployment are not fully accounted.¹ Moreover, in high human-development index countries, the rapid increase of population segments aged 65 and older pose a major challenge in keeping up with the blood supply, since these patients consume multifold red blood count (RBC) units per capita compared to younger patient segments. At the same time, the donor base shrinks in relative terms and, in some countries, absolute terms.^{2,3} It is remarkable, therefore, that blood use in the United States has been relatively stable at approximately 14 million units of blood a year as the population increases in numbers and age.

The present stability of blood consumption is attributed to an increasingly conservative approach in prescribing blood transfusion, both in terms of lowered threshold of blood hemoglobin at which transfusion is deemed necessary (ie, lowering of the transfusion trigger) and the number of units being transfused. Additional factors supporting this trend are preoperative anemia management, as well as anesthetic and surgical modalities that minimize perioperative blood loss. This so-called patient blood management (PBM) approach^{4,5} is also supported by advances in minimally invasive, or laparoscopic⁶ and robotic,⁷ surgery. As a consequence of these developments between 2008 and 2011, the number of blood units transfused in the United States declined by 8.2%, and the available blood supply exceeded demand by 725 000 units.⁸

Nevertheless, even when we consider probable further reductions in the transfusion trigger and PBM treatment modalities, there is a limit of how much blood demand can be reduced

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since the population continues to grow, unless RBC transfusion itself could be replaced by an efficacious blood substitute to fill this unmet need remains a worthy endeavor.

OBJECTIVES IN BLOOD SUBSTITUTE DEVELOPMENT

In principle, blood substitute development and blood transfusion objectives should be intertwined; thus, formulation of a blood substitute requires a specific understanding of the purpose of a blood transfusion and whether it is fulfilled.

In most circumstances, blood transfusion is used to correct decreased circulatory oxygen-carrying capacity. Therefore, it's important to determine how these factors are correlated in everyday medical practice. It is well established that in most cases medical practice errs in favor of safety to prevent hypoxia in vital organs. This leads to a primarily prophylactic use of blood administered in small quantities, which objectively can be shown to have little effect on the required oxygen-carrying capacity.

Pervasiveness of blood transfusion use in small quantities is amply documented. A typical study of the use of blood for intraoperative transfusions in a US hospital shows that 76.0% of patients were transfused with 1 to 3 units of blood, accounting for 44.2% of all the blood used.⁹ Relating these numbers with the normal circulatory oxygen-carrying capacity, it is apparent that a large proportion of transfusion is practiced when blood's oxygen-carrying capacity, although diminished, is more than adequate by a factor of 2. This consideration suggests that globally more than half of the world blood supply is transfused when other, non-blood-dependent approaches could be used.

PHYSIOLOGICAL EFFECTS OF REDUCING BLOOD OXYGEN-CARRYING CAPACITY

There is a tendency to assume that blood oxygen-carrying capacity, determined by the hemoglobin (Hb) content of blood (red blood cell count, hematocrit), is directly related to the ability of the circulation to satisfy the oxygen metabolic demand of the organism; however, this relation is indirect and, in some cases, negative. Initial reductions of hemoglobin, as much as 10% to 20% from normal, in normovolemic conditions, as in clinical anemia, or blood losses corrected with volume-restoring plasma expanders tend to increase cardiac output, tissue blood flow, and oxygen delivery since the consequent hemodilution significantly reduces blood viscosity and peripheral vascular resistance.¹⁰

This example serves to illustrate that the critical factor is not the intrinsic oxygen carrying capacity of blood, but oxygen delivery capacity, namely the product of blood oxygen-carrying capacity and blood flow. This product results in a rate of oxygen delivery that must match or exceed the rate of metabolic oxygen use. Therefore, the critical and real goal of blood transfusion is the restoration and at least equalization of 2 rates: oxygen delivery and metabolic oxygen use. This is a complex problem in which immediate formulation and solution in emergencies transcends conventional, established medical practice. Furthermore, it requires inclusion of the all-important role of blood-flow management into the transfusion paradigm.

Deficiency in oxygen-carrying capacity becomes lethal in extreme anemia; however, in principle, the organism has a sufficient oxygen-carrying capacity margin that allows for surviving the loss of at least half of blood hemoglobin. The problem is that before arriving at this condition, the organism experiences a decrease in oxygen-delivery capacity, as a result of the malfunction of the microvascular perfusion manifested as the fall of functional capillary density (FCD), ie, the number of capillaries per unit volume of tissue with transit of red blood cells.¹¹

FCD has been shown to be the primary determinant of survival following hemorrhage and extreme anemia in experimental studies,¹² independently of tissue oxygen tension, primarily because it ensures extracting the toxic products of metabolism from the tissue. These results highlight the concept that manipulation of a non-oxygen-related variable, microcirculation functionality, can be a means to achieve oxygen delivery in anemia.

However, there is clearly a limit in the absolute reduction of intrinsic oxygen-carrying capacity that can be tolerated since there is a minimal oxygen supply that is needed by the heart muscle to produce flow. When this limit is reached, blood is transfused, although a suitable and efficient nonblood oxygen carrier could serve the same purpose.

IS BLOOD THE OPTIMAL OXYGEN CARRIER?

Blood is the result of an evolutionary process that has maximized the transport of oxygen from the atmosphere to the tissue. In blood, this is optimized by the reversible oxygen binding with the hemoglobin molecule, which is driven by the oxygen gradient between the molecule and its surroundings. High concentrations of hemoglobin molecules are packaged in red blood cells, limiting the exposure of the organism to the free hemoglobin. The elegant design of the system leaves blood as the unchallenged oxygen carrier for the native host organism.

In general, it is shown that blood transfusions are associated with increased bleeding and decreased survival¹³ of acute coronary syndrome patients.¹⁴ An example of the deleterious effect of blood transfusion is the long-term survival of patients after myocardial infarction in which transfused anemic patients had a 50% greater incidence of mortality in the following 6 to 48 months than those who avoided transfusion.¹⁵

The lethality of transfusion is associated with predisposition to postoperative infection,¹⁶ duration of RBC increased predisposition to malignancy,¹⁷ promotion of RBC adhesion to pulmonary endothelium,¹⁸ and reducing of tissue oxygenation due to RBC storage damage¹⁹ to cite a few. The statement “current blood banking storage practices may not adequately protect patients” was the conclusion of a meta-analysis of the relation between mortality and transfusion of stored RBC.²⁰

ALTERNATIVES TO BLOOD TRANSFUSION OF 1 TO 3 UNITS

The preceding considerations apply not only to massive blood transfusion, but are already present with the transfusion of a single unit of blood, andacerbate with increasing numbers. Given the hypothetical physiological alternatives for improving oxygen delivery capacity,

particularly in interventions associated with the pervasive use of 1 to 3 units of blood, there are currently the following potential alternatives to using blood.

Increasing Oxygen-Carrying Capacity With Fluorocarbons

Fluorocarbons have been tested extensively in the clinic and 1 formulation/application received US Food and Drug Administration approval. They ultimately failed in the United States because of minor side effects and the inability to show superiority to blood. A factor is that they were tested and proposed for a market where they are not needed, with an incomplete understanding of how to use their special properties to deliver oxygen to the tissue. Notably, fluorocarbons are universal gas carriers, including volatile anesthetics and nitric oxide, a potentially highly beneficial property that after 3 decades of development is now beginning to be explored.^{21,22} An important aspect that with few notable exceptions is common to all oxygen carriers' development is the lack of physiological studies at the level of the microcirculation, a problem that is currently being corrected²³ for fluorocarbons, and that could have prevented many failures.

Fluorocarbons, at inspired air conditions (21%) and physiological dosages, carry about twice the amount of oxygen of plasma and thus are usually administered in conjunction with hyperoxic ventilation, a procedure that is not yet well characterized in terms of benefit versus potential toxicities. Fluorocarbon development is being pursued in the United States and abroad, and its clinical use is approved in Mexico and Russia.

Increasing Oxygen-Carrying Capacity With Modified Hemoglobins

To date, hemoglobin is unsurpassed as an oxygen carrier. Furthermore, the molecule elicits immune reactions and can be used across species. It is abundant, considering the availability and the suitability of bovine hemoglobin. However, the molecule must be chemically modified, otherwise its tetrameric structure breaks into monomers and dimers that when dissolved in plasma affect kidney function. When this requirement was recognized, many strategies were implemented to ensure the integrity of the tetramer in solution, and several companies were formed to produce an oxygen-carrying solution with properties similar to blood.

The initial efforts to formulate a blood substitute were substantial, having absorbed capital in excess of \$1 billion. The molecules produced, using different chemistry modifications for ensuring the integrity of hemoglobin in solution, have similar biophysical and oxygen transport properties. These molecules underwent clinical trials in the United States and Europe but were not approved because they did not satisfy safety requirements. There is ample literature that analyzes these products and the causes for their lack of success.²⁴

All modified molecular hemoglobin products configured as a small molecule are low-viscosity solutions that scavenge nitric oxide (NO),²⁵ thereby causing systemic hypertension, which is assumed to be a cause for the negative results of clinical trials with these materials. However, it is questionable whether this outcome is directly due to this pressor effect. Hemorrhage is treated with vasopressors such as noradrenaline or arginine-vasopressin,^{26,27} and reports indicate that the hemoglobin pressor effect is beneficial^{28,29} in the management of hemorrhage, providing immediate recovery.³⁰ Hypertension has been

associated with cross-linked hemoglobin solutions, which result in focal necrosis as the result of vasoconstriction,³¹ as well as other medications that similarly impact NO levels.

Oxygen-carrying blood substitutes based on molecular hemoglobin in solution in plasma have persistently yielded negative results in clinical trials because of their inherent toxicity; taming this toxicity has been a major goal of blood substitutes development.^{30,32} This was partially accomplished by conjugation of Hb with polyethylene glycol (PEG-Hb), which was developed commercially by Sangart, Inc. The material is vasoinactive,³³ but it delivers limited amounts of oxygen in clinically relevant conditions. The material was subjected to extensive clinical trials in Europe, but was not approved because it did not demonstrate superiority to blood.

A recent review by Chen et al concluded: “Published animal studies and clinical trials carried out in a perioperative setting have demonstrated that these products successfully transport and deliver oxygen, but all may induce hypertension and lead to unexpectedly low cardiac outputs.”³⁴ Overall, the studies suggest that HBOCs resulted in only modest blood saving during and after surgery, no improvement in mortality, and an increased incidence of adverse reactions.” The meta-analysis of Natanson et al³⁵ found that the clinical trials associated with blood substitutes increased the risk of mortality by 30% and myocardial infarction by 2.7-fold.

Non–Oxygen-Carrying Blood Substitutes

The preceding, albeit limited discussion indicates that restoration of oxygen-carrying capacity as a means of restoring oxygen delivery capacity with non–blood-based transfusion has not been achieved. Furthermore, the development of a toxicity-free oxygen carrier with oxygen transport properties comparable to blood does not appear to be available in the near future. However, as previously discussed, the ultimate goal of increasing oxygen-delivery capacity is in principle achievable by addressing the non–oxygen-carrying capacity component of the oxygen delivery paradigm.

A family of products has become available for increased oxygen delivery capacity based on so-called active plasma expanders. These products were discovered when it was realized that a principal negative component of anemia is the decrease of blood viscosity and that its restoration reestablishes microvascular function, ie, FCD, in the absence of restoration of blood’s oxygen carrying capacity.³⁶ This approach is particularly effective in extreme anemia or the limit where oxygen delivery capacity just matches the metabolic oxygen demand.

High-viscosity plasma expanders, such as Dextran 500 kDa and alginate solutions, were the active plasma expanders that exhibited the capacity of significantly improving oxygen-delivery capacity in extreme anemia and hemorrhage.³⁷ An unexpected property of this form of plasma expansion was the significant increase of blood flow and tissue perfusion, a phenomenon that led to the labels “supra-plasma expansion” and “supra perfusion.”

These phenomena were extensively studied at the University of California, San Diego, leading to the finding that polyethylene glycol conjugated albumin (PEG-Alb) is an optimal

supra-plasma expander, because its viscogenicity is shear rate dependent, ie, its viscosity is high in the low shear stress segments of the circulation and vice versa.³⁸ This mechanism becomes engaged when PEG-Alb is infused in lieu of a 1- to 2-unit blood transfusion. Cardiac output and blood flow are augmented because the increased viscosity in the low shear stress regions of the circulation, mostly in the microcirculation, induces the production of the vasodilator NO, while facilitating flow in the high shear regions. The net effect is that heart workload is decreased and vascular flow resistance is decreased and supra perfusion is produced.³⁹ Increased NO production by mechanotransduction due to increased microvascular wall vessel shear stress due to presence of the plasma expander provides the additional benefit of lowering the oxygen.^{40,41}

The benefits of supra-perfusion-inducing plasma expanders extend beyond the increase of oxygen-delivery capacity. Blood transfusion is preceded by an anemic phase caused by hemodilution, which results from the use of conventional plasma expander with the aim to maintain volume. This phase introduces changes in the mechanical and chemical environment of the endothelium, which propagate on transfusion. The changes include activation of genetically controlled mechanism, such as endothelial impairment due to inflammatory reactions;⁴² activation of endothelium, platelets, and neutrophils; and liberation of cytokines. At the cellular level there may be endothelial swelling and increased endothelial permeability due to ischemic injury.⁴³ Initial plasma expansion with PEG-Alb has demonstrated significantly better results following the blood transfusion phase in terms of ultimate recovery of microvascular function and survival.⁴⁴ Since these fluids increase blood flow due to reduced blood viscosity and vasodilation, they are effective at lower blood pressures diminishing bleeding and interference with clot formation. In addition, dilutional coagulopathy may be minimized with supra-perfusion-inducing plasma expanders, which do not require the large infusion volume of crystalloids/colloids. However, the main administration challenge will still be to ascertain accurately how close the patient is to the oxygen supply limit.

CONCLUSIONS

RBC transfusion unquestionably has the potential to secure immediate and sometimes long-term survival when used for acutely restoring oxygen-carrying and -delivery capacity following surgery and accidents. However, it ignores the longer range effect on mortality due to transfusion, abundantly reported in the literature, showing that transfusion of even 1 unit of blood following surgery increases mortality by 10% over no blood transfusion after 10 years. This level of reliability (ie, 1 failure in 10 at 10 years posttransfusion) shows there should be a large demand for a blood delivery restorer by populations whose economic conditions can afford the potentially high cost of a nonblood product. In addition, risks of iron overload from chronic transfusion therapy could be reduced with the use of interventions promoting perfusion leading to a better management of the ability of circulating red blood cells to delivery oxygen, thus decreasing total body iron and the risk of long-term iron-related sequelae.⁴⁵

This situation applies to small- and large-unit transfusions of blood and is particularly important in the small-volume intervention, since this relates to close to half the transfused

blood supply and the transfused population. Large-scale data analyses show that this type of transfusion is, in principle, avoidable. Furthermore, when it is necessary, a new nonblood, non-oxygen-carrier approach with apparently no known toxicity (based on the induction of the supra-perfusion effect) is potentially available and endowed with additional benefits. Inducing supra perfusion has significant additional medical benefits of enhancing the removal of metabolic waste products from the tissue, the standard treatment for endotoxemia and septic shock.

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Biographies

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