Factors Affecting Tissue Oxygenation in Erythrocyte Transfusions

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Red blood cell transfusions are used to increase the oxygen-carrying capacity of blood in anemic states. But, because of the changes during storage of blood components and the specifics of preparation, erythrocytes may have controversial effects on tissue oxygenation and microcirculation. Also, the patient situation may play a role in the differing responses in oxygenation and microcirculation. In this review, the studies concerning the effects of banked blood and patient characteristics on microcirculation and tissue oxygenation are summarized.

Key Words: Erythrocytes, blood transfusion, microcirculation

Introduction

The main purpose of red blood cell transfusion is to provide oxygen-loaded blood to the microcirculation in order to maintain tissue oxygenation in anaemia (1). However, the studies shows that it is not always possible. Numerous clinical studies on this subject have demonstrated that complications such as mortality, organ failure, infection and prolonged hospital stay increases in proportion to blood storage duration. Besides, there are studies showing that storage duration has no effect on complications. However, current research on blood transfusion point out to the role of the quality of blood transfused to patients. As is known, erythrocytes go through several biochemical and hemorheological changes during storage. In many clinical studies, the results of these reactions have been reported to affect blood quality thereby leading to transfusion complications. Additionally, there have been remarks on the effects of leukocytes in stored blood (2, 3). The effects of patient characteristics on erythrocyte transfusion should also not be forgotten (4, 5). There are conditions that tissue oxygenation cannot be maintained despite improvement in the haematocrit levels of the patient after transfusion. This situation is more commonly encountered during severe infections such as sepsis (6). Although the studies of our group on sepsis patients have yielded the same results, clinical studies we performed on cardiac and haematological patients showed that transfusion has a beneficial effect on tissue perfusion and oxygenation (6-9).

In conclusion, the majority of clinical and experimental studies on blood transfusion have evaluated the effect of factors related to blood storage on transfusion, and patient characteristics (disease and disease severity, the degree of anaemia, treatment plan) have not been taken into account. In this report, after a short history of blood transfusion, we will review the studies on blood transfusion with regard to factors related to blood storage as well patient characteristics.

History

William Harvey proposed his theory on blood circulation in 1628; however, our knowledge on blood transfusion is based on earlier observations. The first known blood transfusion was performed in 1490 to Pope Innocent VIII. It was reported that blood of three children at 10 years of age was used to rescue the Pope (10). Animal-to-animal blood transfusion was first attempted in 1665 by Richard Lower and Jean-Baptiste Dennis, who succeeded in their attempts in 1667. The subsequent important step in blood transfusion was taken by the English gynaecologist James Blundel. Blundel was the first performing human to human blood transfusion in 1818. Thereafter, between 1818 and 1829, four of ten attempts using human blood for transfusion, succeeded. Beginning from this period, blood transfusion has carried the risk of haemolytic

transfusion reactions until the 20th century. The discovery of ABO blood groups in 1901 by Karl Landsteiner, and thereafter usage of sodium citrate as an anticoagulant solution in 1915 by Richard Lewison are very important steps in transfusion history. Furthermore, Karl Landsteiner with Alexander Wiener described Rh blood group system in 1940. Besides, the importance of blood transfusion was better understood during the world wars, and it triggered the research on colloids and plasma. In the last 30 years, studies about the changes in the properties of blood associated with storage come to the forefront.

The Effects of Changes in the Properties of Blood Associated with Storage Duration on Tissue Oxygenation Following Transfusion

Review of research on biochemical and hemorheological changes

The changes in biochemical and hemorheological properties of erythrocytes (11) during blood storage comprise an important cause of post-transfusion problems. Biochemical changes include decreased 2-3 DPG and ATP levels (12), loss of membrane sialic acid (13), impairment of lipid peroxidation (14), loss of intrinsic membrane proteins (15), loss of antioxidant capacity (16), decreased pH (17), increase in free haemoglobin concentrations due to haemolysis (18), and reduced S-nitroso-haemoglobin levels (19). Besides, the changes in the hemorheological properties of erythrocytes result in easy deformation (20, 21), aggregation and increased adhesion (22).

Among the above-mentioned biochemical changes, the most important problem related to the transfusion of stored blood is decrease in 2-3 DPG levels. As known, decrease in 2-3 DPG levels will slow down the release of oxygen to the tissues by erythrocytes. However, this slowing down can be reversed to its previous state in 72 hours (23). The decrease in ATP levels may lead to important consequences such as decreased erythrocyte survival. We, in a study of our group on ATP levels, proved that the decrease in ATP levels during erythrocyte storage could be reversed by a special solution, providing an improvement in the renal microcirculation of rats (24).

Increase in free haemoglobin due to haemolysis during storage also leads to important consequences. The study of Donadee, published in 2011, is very interesting. Donadee, in her research, showed that free haemoglobin released from erythrocytes during blood storage, increased the consumption of nitric acid more potently than normal erythrocytes (18) and she highlighted the vasoconstrictor properties of free haemoglobin.

The adhesion and aggregation tendency of erythrocytes due to changes in hemorheological properties during storage prevents the entry of cells into the microcirculation after transfusion (2).

In conclusion, biochemical and hemorheological changes during storage are among the important factors affecting tissue oxygenation following transfusion.

The review of research on leukocytes

Leukocytes are also considered among the factors ruining the positive effects of blood transfusion on tissue oxygenation. It is thought that cytokines, enzymes and inflammatory mediators released from leukocytes are responsible for this condition. Leukocyte filtration that is performed to decrease the immunosuppressive effects of blood transfusion is an issue open to question (25, 26). Anniss and colleagues in their study on the adhesive properties of erythrocytes in leukofiltered blood demonstrated that leukoreduced blood showed less adhesion, and had a positive effect on microcirculation. Another researcher evaluating the effects of leukocyte filtration was Van de Watering. Van de Watering demonstrated that the survival rate of cardiac patients receiving leukocyte-filtered blood was higher than the patients receiving buffy-coat-depleted red blood cells (28). Netzer et al. (29) also showed that the mortality rate of patients receiving leukoreduced blood was lower in comparison to that of patients receiving non-leukoreduced blood. However, another study carried out on patients who were given leukocyte-depleted blood yielded controversial results. Dzik, in a randomized prospective study on 2780 patients found that white blood cell-reduced blood had no effect on mortality rates (30). Similarly, Nathens, in a randomized study on 1864 trauma patients claimed that mortality and disease rates did not change in patients receiving leukoreduced blood (31).

Experimental research on the subject

Multiple experimental studies performed on animal models of anaemia, document that the effects of fresh blood transfusion are much more better than stored blood on microcirculation. For instance, Van Bommel in a study on rats showed that fresh blood transfusion after haemorrhage had a more favourable effect on the intestinal microcirculatory oxygenation than stored blood (20). In this study where no difference was detected between the groups in terms of venous oxygen saturation, Van Bommel additionally pointed out that different hemorheological changes produced by different solutions used in blood storage have differing effects on intestinal microcirculatory oxygenation.

Also, Tsai and colleagues in their research showed that hemodilution induced hamsters, demonstrate a significantly malperfused microvasculature (32). However, as the effect at skin level was not detected at the systemic level, remarks about microcirculation are not definitive. Additionally, as the stored blood (for 28 days) was not leukoreduced, the results of this study are debatable.

d'Almeida and colleagues compared the biochemical changes in stored blood samples obtained from human and rat donors and observed that rat erythrocytes stored for one week develop a storage lesion similar to that of human blood stored

for 4 weeks (33). The researchers indicated that there may be significant differences in the structure and metabolism of erythrocytes between species.

Raat et al. (12), who planned to conduct a clinically relevant study, produced a rat model that can tolerate human blood and evaluated the effects of human blood on rat intestinal micro-circulation. They demonstrated that oxygen delivering capacity of blood stored for 6 weeks was lower than that of fresh blood.

The relevant clinical studies

Numerous clinical studies show that increased blood storage duration trigger complications like post-transfusion death (34, 35), organ failure (36) and prolonged hospital stay (37, 38). However, there are studies showing no effect of transfusion of stored blood (39-42).

The studies which reported complications due to transfusion of blood stored for long periods are listed in Table 1. Purdy and colleagues observed that mortality rates were higher in septic patients who received stored blood (34). Vamvakas, in another retrospective study on 416 cardiac patients, showed that risk of postoperative pneumonia increased in patients receiving stored blood (37). A study on this topic was carried out in trauma patients by Zallen et al. (36) Zallen retrospectively evaluated the effects of blood transfused in the first 12 hours of admission to the trauma centre, and proved that organ failure rate was higher in patients who were transfused blood stored for more than one month. Offner, in another study performed in the same centre on trauma patients showed increased infection risk and prolonged intensive care stay in patients who received stored blood (38). However, contrary to the study of Zallen, Offner did not observe an increase in the mortality and organ failure rates of patients who were given stored blood. In another study on trauma patients, Weinberg and colleagues evaluated the mortality rates of 1813 trauma patients after blood transfusion. Weinberg included patients who received more than one unit of blood in the first 24 hours of admission to the trauma centre, and observed that mortality risk increased with volume and that

transfusion of smaller volumes of stored blood had no effect on mortality (43).

Another study, assessing the effects of stored blood was performed by Koch et al. (35) Koch studied cardiac patients in two groups; the first group received blood that was stored for 14 days or less and the second group received blood stored for more than 14 days. In- and out-of-hospital mortality and postoperative complications increased in patients receiving older units (35). However, there are two controversial issues in this study. Firstly, the maximum permitted storage duration in the USA is longer than that in Europe; therefore, the results of this study should be compared with caution. On the other hand, although the complications were found to be increased in proportion to the blood storage duration, it should be considered that more than half of the blood used was non-leukoreduced blood.

The studies that reported no complications due to transfusion of stored blood are presented in Table 2. Vamvakas, in a study in 1999, observed an increase in postoperative pneumonia risk in cardiac patients who received stored blood, and in another study on the same patients showed that blood storage duration did not have an effect on the length of intensive care and hospital stay (39). In another study on this issue Walsh evaluated septic patients, and randomized those who were transfused with blood stored for more than twenty days to the first group, and the patients who received blood stored for less than 5 days to the second group. Walsh could not detect any significant difference between these two groups (40). A relevant study on trauma patients was carried out by Murrell and colleagues. Murrell made a retrospective research on 275 trauma patients, and found no change in the mortality rate of patients who received stored blood. However, the length of intensive care unit stay was significantly longer (41). van de Watering who performed another study on the same topic in 2732 cardiac patients, found that blood storage duration had no effect on mortality and length of intensive care unit stay (42). However, it should be kept in mind that the blood transfused to the patients in that study was non-leukoreduced blood.

Table 1. Clinical studies reporting complications after transfusion depending on duration of blood storage

Author	Publishing year	Patient group	Complication following transfusion
Marik (4)	1993	23 septic patients	Gastric pCO ₂
Purdy (34)	1997	31 septic patients	Mortality
Vamvakas (37)	1999	416 cardiac surgery patients	Postoperative pneumonia
Zallen (36)	1999	63 trauma patients	Organ failure
Offner (38)	2002	61 trauma patients	Length of intensive care stay infections
Weinberg (43)	2008	1813 trauma patients	Mortality Renal failure Pneumonia
Koch (35)	2008	6002 cardiac surgery patients	Mortality Postoperative complication

Table 2. Clinical studies with no observed complications due to blood storage duration after transfusion

Author	Publishing year	Patient group	Parameters
Vamvakas (39)	2000	268 cardiac surgery patients	Length of intensive care stay postoperative pneumonia
Walsh (40)	2004	22 septic shock patients	Gastric tissue oxygenation
Murrell (41)	2005	275 trauma patients	mortality
Van de Watering (42)	2006	2732 cardiac surgery patients	mortality Length of intensive care stay
Yürük (8)	2011	20 haematology patients	Microvascular density Microvascular oxygenation

Table 3. Clinical studies evaluating the direct effects of erythrocyte transfusion on microcirculation

Author	Publishing year	Patient group	The effect on microcirculation
Genzel-Boroviczeny (46)	2004	13 preterm paediatric patients	Increase in skin capillary density
Sakr (44)	2007	35 septic patients	No effect was observed on sublingual microcirculation
Creteur (45)	2009	44 intensive care patients (18 septic patients)	No effect was observed on muscle tissue oxygenation, oxygen consumption, microvascular reactivity
Yürük (7)	2011	24 cardiac surgery patients	Increase in sublingual microvascular density and tissue oxygenation
Yürük (48)	2012	20 haematology patients	Increase in sublingual and muscle tissue oxygenation

Due to reasons as such, the results of studies performed in centres implementing transfusion policies other than current implementations and the results of the majority of the above-mentioned studies should be interpreted very carefully. Another study setting a good example for this condition is the study of Sakr et al. Sakr observed in the retrospective analysis of intensive care patients that increased haemoglobin concentration after blood transfusion decreased hospital deaths in postoperative intensive care patients (except those who had undergone cardiac surgery) and in severe disease groups such as sepsis (6). The results of this study performed by Sakr and colleagues are debatable.

Another study that showed no effect of storage duration on tissue oxygenation was performed by our group (8, 12). In that prospective randomized clinical study, 20 haematology out-patients were evaluated in two groups. The patients who had received leukoreduced blood stored for less than 1 week were randomized to the first group, and those who had received leukoreduced blood stored for 3-4 weeks were randomized to the second group. Although there was a parallel increase in haemoglobin values, oxygen saturation and perfused capillary density, there was no significant difference between the two groups.

The Effects of Patient Characteristics on Tissue Oxygenation in Erythrocyte Transfusions

The effects of pre-existing diseases

As mentioned above, studies examining the effects of blood transfusion on tissue oxygenation mostly highlight the results of changes associated with the storage duration of the blood

used for transfusion. However, patient characteristics should also be considered among factors affecting tissue oxygenation. The simplest example of this is seen in severe disease conditions like sepsis. Studies performed on sepsis patients reveal that blood transfusions do not produce a treatment effect on microcirculation (2, 4). Among its reasons, mediators of inflammation have been shown to further increase microvascular obstruction and shunts in sepsis patients.

Studies on the direct effects of erythrocyte transfusion on microcirculation based on patient characteristics are very few (7, 8, 44-46). These studies that are presented in Table 3, indicate that the results differ depending on the patient group analysed. While microcirculation is not affected by transfusion in septic patients (44, 45) due to endothelial dysfunction (47, 48), transfusion has a favourable effect on microcirculation in adult cardiac patients (7), adult haematology patients (8) and preterm paediatric (46) patient groups.

The first study on sepsis patients was performed by Sakr et al. Sakr and colleagues investigated the effects of transfusion on sublingual microcirculation by using OPS (Orthogonal Polarization Spectral imaging) in 35 sepsis patients (44). Study measurements were scheduled to be made immediately before transfusion and one hour after transfusion. The blood that was used for transfusion was leukoreduced blood stored for less than 24 days. Sakr et al. (45) could not detect any changes in post-transfusion oxygen intake and microcirculation parameters although there was an increase in blood pressure and the rate of oxygen delivered to the tissues. Another study on sepsis patients was performed by Creteur and col-

leagues. Creteur also found similar results to the Sakr study and determined no difference in muscle tissue oxygenation, oxygen consumption and microvascular reactivity parameters in sepsis patients after transfusion of leukoreduced blood. The mean storage duration of blood was 18 days in the study of Creteur. Another study of our group on sepsis patients examining the effects of erythrocyte transfusion on microcirculation yielded supportive results (9).

Different from the above mentioned two studies, the most important study showing the beneficial effects of erythrocyte transfusion on microcirculation is the study by Genzel-Boroviczeny. Genzel-Boroviczeny observed an increase in skin capillary density of preterm paediatric patients at 2 and 24 hours after transfusion (46). Similar results to the study of Genzel-Boroviczeny have also been observed by our group. In the study of our group on adult cardiac patients, an increase was observed in sublingual microvascular density and tissue oxygenation parameters after transfusion (7). Besides, our study on adult haematology patients also showed an increase in sublingual tissue and muscle oxygenation following transfusion (8, 49). These two studies performed by our group were able to show that erythrocyte transfusions improved tissue oxygenation by affecting microcirculation. The underlying mechanism is the perfusion of previously closed capillaries after transfusion. There is another study demonstrating the favourable effects of erythrocyte transfusion on microcirculation. That study performed by our group on a surgical patient group verified that fresh non-leukoreduced RBC transfusions were more effective in improving sublingual microcirculation than stored blood (50).

Conclusion

The main purpose of erythrocyte transfusion is providing oxygen loaded blood to microcirculation to improve tissue oxygenation in anaemia. However, studies evaluating the direct effects of erythrocyte transfusion on microcirculation are very rare. Therefore, as the first step, clinical studies investigating the effects of blood transfusion at the microcirculatory level should be planned. Besides, it is also important to explore new ways to reopen the capillaries that are resistant to erythrocyte transfusions in sepsis patients. For this purpose, first of all the mechanism of capillary resistance in sepsis patients should be enlightened. Only by this way will we achieve favourable results with erythrocyte transfusions in patients with sepsis or other disease conditions.

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