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Role of erythropoietin in the newborn

Since the isolation of the human erythropoietin gene in 1985[1] there has been interest in the possible use of recombinant human erythropoietin (r-HuEpo) as an alternative treatment to blood transfusion in preterm infants. Several studies have now been published reporting varying degrees of response but as yet no conclusive evidence has been presented to support the routine use of r-HuEpo in the preterm infant.

Current need for transfusion

The requirement for blood transfusion for preterm infants of less than 1500 g birth weight is well recognised.[2] There are two main groups of preterm infants who require blood transfusion: the first being those who require early transfusion during the first few weeks after birth and the second those who develop anaemia at around six weeks after birth the so called (early) anaemia of prematurity. A late anaemia of prematurity occurring after several months is almost entirely due to iron deficiency and will not be addressed further here.

The first group of sick, often ventilated, preterm infants requiring intensive care receive the majority of blood transfusions. These preterm infants are one of the most frequently transfused groups of patients receiving a mean of four (range 0–10) transfusions during the first 28 days of life.[2] Although there are many causes for the development of this early anaemia, the main aetiological factor is the need for multiple blood tests for intensive care management. It has been shown that up to 67 ml/kg of blood may be removed during the first four weeks of a preterm infant's life.[2–4] These early transfusions accounting for the majority of the blood transfusions given to preterm infants[5] and are very unlikely to be ameliorated by r-HuEpo.

The second group of infants require blood transfusion at around 6 weeks of age because of the anaemia of prematurity but are often otherwise healthy. The anaemia of prematurity is an exaggeration of the fall in haemoglobin that all infants undergo during the first months of life.[6] With infants of earlier gestation the anaemia of prematurity is more profound. These infants often develop signs of anaemia and require transfusion. The aetiology of the anaemia of prematurity is in part related to the universally low serum erythropoietin concentration (with associated low reticulocyte counts) found even in the presence of anaemia.[7–10] It is the anaemia of prematurity that has been the target of the currently published clinical studies into the role of erythropoietin in preterm infants.

Risks associated with blood transfusion

Whether given in the first few weeks after birth or later for the anaemia of prematurity, the use of blood products in preterm infants continues to be of concern due to the risks associated with transfusions. The main anxiety is the significant risk of transmission of viral agents through blood products. Until recently the most frequent viral infection transmitted was cytomegalovirus and, along with hepatitis B and C, transmission of cytomegalovirus continues to be a small but significant risk.[5] Before the routine use of cytomegalovirus negative blood products for preterm infants there was a significant infection rate of 25–30% associated with cytomegalovirus positive blood, with a mortality of around 25% of those infected.[11] This has been substantially reduced by the use of cytomegalovirus negative blood in preterm infants.

The risk of HIV transmission by blood transfusion in the UK is currently estimated to be less than one in a million transfusions,[12] although up to 20/million in parts of the USA[13] and considerably higher in other parts of the world.[14] One of the earliest reports of transfusion associated HIV infection arose in an 18 month child who was repeatedly transfused at birth.[15] There continues to be concern that there could be another yet unknown transmissible agent with the devastating effects of HIV around the corner. The risks are increased by multiple transfusions from many different donors and can be lessened by the repeated use of blood from a single donor unit for an individual infant.

Many parents of preterm infants express natural concerns over the safety of transfusions, and the specific religious objections from Jehovah's Witnesses and other groups also pose difficult problems.[16] With this background, several studies have now been published that seek to address the question of the efficacy of r-HuEpo in the preterm infant.

Evidence for a biological response to r-HuEpo

Several in vitro studies using cell culture techniques have demonstrated that preterm infants with the anaemia of prematurity born between 27–33 weeks' gestation have adequate numbers of erythroid progenitors.[17–19] The progenitors from both peripheral blood[17 18] and bone marrow[19] are responsive to r-HuEpo in vitro.

A wide range of doses of r-HuEpo has been tried in the limited number of clinical studies so far published, ranging from 70 U/kg/week[20] to 1200 U/kg/week.[21] Some of these

studies have been preventive, with r-HuEpo treatment commencing on the second,[21] third,[20] or eighth day[22] after birth without any additional packed cell volume criterion. Other studies have sought to treat the anaemia of prematurity by commencing r-HuEpo treatment around three,[23 24] four,[25 26] or six[27] weeks after birth, when the packed cell volume is below a defined level. This difference in the fundamental design of the studies has reduced the comparability of the results.

Using the reticulocyte count as a measure of the response to r-HuEpo, there was no significant difference between the treated and control groups in a double blind study of 10 infants given 200 U/kg/week r-HuEpo intravenously from 3 weeks of age.[24] However, in a historically controlled study using doses ranging between 75–600 U/kg/week[23] from 4 weeks of age, a twofold increase or greater was seen in all but one of 18 infants, and this appeared to be in a dose dependent fashion. A further double blind randomised study using doses between 100–300 U/kg/week given subcutaneously from 8 days of age, demonstrated a sustained and significantly elevated mean reticulocyte count of 110×10^9/l throughout the six week study period in treated infants (n=15) compared with 55×10^9/l ($p<0.05$) in the placebo group (n=8).[22]

Other studies have employed significantly higher doses. One study giving between 500–1000 U/kg/week r-HuEpo subcutaneously to just four infants from 3 weeks of age showed a significantly increased mean reticulocyte count of 262×10^9/l compared with 136×10^9/l in the placebo group.[25] A similar marked rise in the reticulocyte count to 7% from a pretreatment level of 2% was found in a study comparing 700 U/kg/week r-HuEpo treatment subcutaneously (n=10) with transfusion (n=9) beginning at 6 weeks of age.[27] A marked reticulocyte response was also seen with early r-HuEpo treatment from 2 days of age in a study using a high dose up to 1200 U/kg/week of 4·46% in the r-HuEpo group (n=11) compared with 1·1% for the controls ($p=0.0001$).[21] These results suggest that r-HuEpo will stimulate a reticulocyte response in a probable dose dependent fashion whether given early or late, and that this will be sustained throughout the treatment.

Additional evidence for active erythropoiesis in infants treated with r-HuEpo has been demonstrated in several studies by a rapid fall in the ferritin level.[22 23 27] Adequate iron supplies are essential for an optimal response to r-HuEpo in adults with chronic renal failure.[28] There remains debate about the amount of iron supplementation that is required to prevent deficiency developing as a result of r-HuEpo treatment in preterm infants. The amount of supplementary iron in the reported studies has ranged from 2 to 8 mg/kg/day orally[23 26] while one study administered 20 mg/kg of iron intravenously each week to prevent deficiency developing.[21]

Evidence for a reduction in the need for transfusion

The ultimate end point for any study of the use of erythropoietin whether for the treatment or prevention of the anaemia of prematurity, must be a reduction in the number of transfusions given, and few studies have clearly addressed this issue. In one double blind study using 200 U/kg/week intravenously from 3 weeks of age, there was no significant difference in the numbers of infants requiring transfusion in either the r-HuEpo or placebo groups.[24] In a second double blind study using broadly similar doses but with treatment beginning at 8 days of age, there was a 41% reduction in the number of transfusions required, although this too did not reach statistical significance.[22] However, with doses of 1200 U/kg/week of r-HuEpo also given early from 2 days of age (n=11), significantly fewer transfusions were required at 0.8 ± 1.5 compared with 3.1 ± 2.1 without treatment ($p=0.01$).[21] In addition, in this study a smaller volume of packed erythrocytes was transfused in the r-HuEpo group at 14·2 ml/kg compared with 48·4 ml/kg in the control group.[21]

Other studies have been extremely small (n=4)[25] or have not included adequate control groups to enable comparison.[23 26] These studies have used changes in the packed cell volume as a pointer to the efficacy of r-HuEpo but these data remain unconvincing. One study using 75–300 U/kg/week showed a rise of 3·3% (not significant) after three weeks of treatment (n=7),[26] while in a subsequent study by the same group using doses of 75–600 U/kg/week (n=18) there was a smaller rise of 1·1% also at three weeks.[23] In a double blind study using doses of 500–1000 U/kg/week commencing at four weeks after birth (n=4) there was a fall in packed cell volume by 2% in the r-HuEpo treated group compared with a fall of 8·4% in the placebo group ($p=0.0007$) after six weeks of treatment.[25]

As outlined above, the currently published studies have great differences in design and in the response observed to r-HuEpo treatment. No conclusion can yet be drawn as to the optimal dose of r-HuEpo, the age at first dose, the frequency of administration, the route of administration, the length of the treatment phase, and the amount of iron supplementation required. All the studies have small numbers of infants in the treatment arm, ranging from 43[20] to as few as four.[25] With few studies using a double blind randomised design there are still limited data available as to the efficacy of r-HuEpo treatment for either the prevention or the treatment of the anaemia of prematurity.

Other aspects of r-HuEpo treatment

The development of neutropenia in the r-HuEpo treated group has been reported in several uncontrolled studies,[23 26 27] with the neutrophil count falling to a mean value of 0.8×10^9/l at 56 days after commencing treatment, compared with 2.2×10^9/l at the outset in one study.[26] Although other studies have not found a significant fall,[21 22 24 25] an inverse relationship between the neutrophil and the reticulocyte count has been observed.[24] A decreased neutrophil storage pool on tibial bone marrow aspirates taken between seven and 10 days after onset of r-HuEpo treatment has also been noted.[27] Transient small rises in the platelet count have been observed[23 26] and a positive relationship between the reticulocyte count and the platelet count is reported.[24] The significance of these findings is yet to be clarified.

No consistent adverse events have been identified in the published studies, and although two infants in the r-HuEpo treated group of one study died of sudden infant death syndrome four weeks after cessation of r-HuEpo treatment,[22] the relationship of these events to the use of r-HuEpo remains unclear. No deaths have occurred in the studies administering high doses of r-HuEpo.[21 25]

Conclusion

In conclusion, r-HuEpo stimulates reticulocyte response in a probable dose dependent fashion. It would also appear that r-HuEpo given in an adequate dose for a sufficient period of time, with adequate iron supplementation, offers the promise of a reduction in the need for transfusion for the anaemia of prematurity. There are no data as yet to suggest that r-HuEpo will provide an alternative form of treatment for the anaemia of prematurity, but there is some evidence that r-HuEpo commenced shortly after birth may reduce the number of transfusions by preventing

the development of the anaemia of prematurity.

There is a need for large multicentre double blind randomised studies, where significant numbers of infants can be recruited, so as to determine the efficacy and safety of r-HuEpo in reducing the need for transfusion for the anaemia of prematurity. To date, multicentre studies are currently being undertaken in the US and Europe and the results are eagerly awaited. Until these data are available there is little place for further small inadequately controlled studies, or for the *ad hoc* use of r-HuEpo for the treatment or prevention of the anaemia of prematurity.

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