

A Comparative Study of Cord Blood Haemoglobin Levels and Duration of Treatment in Maternal ABO and Rh-D Antibody Mediated Haemolytic Disease of Fetus and Newborn

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ABSTRACT

BACKGROUND

Haemolytic Disease of Fetus and Newborn (HDFN) is characterised by lysis of red blood cells resulting in anaemia and its hypoxic effects thereafter. Following anaemia, the production of fetal red blood cells is drastically increased. This is followed by extramedullary haematopoiesis in a widespread manner and erythroblastosis characterized by nucleated red cells in the circulation. Since this is an illness affecting many a newborn, assessing the patterns of anaemia in both ABO and Rh-D HDFN may help in effective planning and implementation of better management protocols.

METHODS

This is a cross sectional study with comparative analysis. The study was performed among 154 neonates who were diagnosed to have HDFN. Setting for this study was Department of Transfusion Medicine and Pediatrics of Government Medical College, Trivandrum. During the time of delivery, 5 mL umbilical cord blood was collected. That sample was used for analyzing haemoglobin levels of the neonate. Duration of treatment of the neonate was noted down during follow up. Statistical analysis was done using SPSS software version 16.

RESULTS

The mean cord blood haemoglobin value in ABO haemolytic disease was 17.1 ± 2.7 g %. The mean cord blood haemoglobin value in Rh-D haemolytic disease was 14.5 ± 1.9 g %. In infants with ABO haemolytic disease, 53 (48.2 %) had undergone no treatment in ICU. Remaining 51.8 % had undergone 3 - 10 days of treatment in neonatal ICU. Infants with Rh-D HDFN 20 (45.5 %), had undergone no treatment in ICU. Remaining 54.5 % has got 3 - 9 days of treatment in neonatal ICU. Mean treatment duration in infants with ABO haemolytic disease was 3.1 ± 3.3 days. Mean treatment duration in infants with Rh-D haemolytic disease was 2.9 ± 2.9 days. A comparison of mean values of cord blood haemoglobin in neonates with both categories of haemolytic disease was performed. The p value was 0.00 and it was significant. Mean values of duration of treatment of neonates with ABO and Rh-D haemolytic anaemias were compared statistically. No significant difference was observed.

CONCLUSIONS

Mean haemoglobin levels in Rh-D HDFN showed a significant decrease as compared to ABO-HDFN. Duration of treatment in ABO HDFN and Rh-D HDFN presented no significant difference.

KEYWORDS

Haemolytic Disease of Fetus and Newborn, Umbilical Cord Haemoglobin, Anaemia, ICU Stay, ABO HDFN, Rh-D HDFN

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BACKGROUND

HDFN can cause hyperbilirubinaemia and anaemia in neonates. Hypoxic effects of anaemia and toxic consequences of hyperbilirubinaemia can result in deleterious effects to neonatal health.¹ Following anaemia there will be increased red blood cell synthesis. Consequently, it might result in extramedullary haematopoiesis and erythroblastosis. This is usually characterized by nucleated red cells in the neonatal circulation.² Additionally hydrops fetalis might be characterized by severe anaemia, massive oedema, hypoproteinaemia, stillbirth etc.³

In HDFN maternal IgG antibodies are transferred through placenta into neonate. That antibodies can significantly reduce lifespan of neonatal erythrocytes.³ This can be caused by any of the red antibodies. Most common antibodies causing HDFN include anti-A, anti-B and anti-D. In ABO-HDFN, naturally occurring anti-A or anti-B can pass through placenta. While in Rh-D HDFN, immunization occurs due to previous transfusion or pregnancy related events where Rh-D positive cells have entered maternal circulation.²

In 'Kerala model' of health development there is lower rates of infant mortality.⁴ Since HDFN is a condition that can result in infant morbidity and mortality, it was very important to study various aspects of this illness. The research findings can help in effective planning and implementation of better management protocols for HDFN.

Objectives

To compare cord blood haemoglobin levels and duration of treatment in maternal ABO and RH-D antibody mediated haemolytic disease of fetus and newborn.

METHODS

This is a comparative cross sectional study conducted among 154 neonates who were having HDFN. The study subjects were enrolled by random sampling method. Study was initially approved by Human Ethical Committee of the hospital. Study was started after obtaining permission from review board of institution. (IEC no; 02 / 48 / 2012 / MCT / 16 / 02 / 2012) Study was done for a period of 18 months in 2012. Setting for this research was Department of Transfusion Medicine and Paediatrics, Government Medical College, Trivandrum.

Study was performed in neonates who fulfilled the inclusion criteria. Inclusion criteria included for ABO haemolytic disease included hyperbilirubinaemia on day 1, materno-neonatal ABO incompatibility, ABO maternal IgG titre ≥ 32 and neonatal DAT and / or elution positive or presence of maternal antibodies in cord blood serum. For Rh-D haemolytic disease the inclusion criteria were maternal antibody screen positivity for anti-D, absence of materno-neonatal ABO incompatibility, positive direct antiglobulin test and elution in neonate along with hyperbilirubinaemia on day 1. Those neonates suffering from co morbid illnesses

which can result in anaemia or hyperbilirubinaemia was excluded from the study. Parents of all neonates were counselled and consent was obtained.

Demographic details were noted. During the time of delivery, a 5 mL umbilical cord blood was collected. That sample was used for analysing haemoglobin levels of the neonate. Duration of treatment of the neonate was noted during follow up.

Statistical analysis was performed using SPSS software version 16. Expressions of continuous variables were as mean along with standard deviation. The discrete data was analysed as frequencies and percentages. Independent t test was used for comparing mean of two categories. All p values < 0.05 were considered statistically significant.

RESULTS

Parameter	Frequency	Percentage
Hyperbilirubinaemia due to ABO HDFN	110	71.4
Hyperbilirubinaemia due to RH-D HDFN	44	28.6
Total Hyperbilirubinaemia Cases	154	100

Table 1. Distribution of ABO and RH-D HDFN

Among 154 cases of HDFN, 71.4 % (110) was due to ABO antibodies and 28.6 % due to Rh-D antibodies.

Parameter	ABO HDFN (n = 110)	Rh-D HDFN (n = 44)	P-Value [§]
Mean (g %)	17.1 \pm 2.7	14.5 \pm 1.9	0.0001*

Table 2. Comparative Analysis of Cord Blood Haemoglobin Values

* - Indicates significance at $p < 0.05$; § Independent t-test has been applied as the test of significance

The mean cord blood haemoglobin value in ABO haemolytic disease was 17.1 ± 2.7 . The mean cord blood haemoglobin value in Rh-D haemolytic disease was 14.5 ± 1.9 . The median cord blood haemoglobin values in ABO and Rh-D HDFN infants were 17.1 and 13.9 g % respectively. Minimum haemoglobin value in ABO haemolytic disease was 11.1 g % and in Rh-D haemolytic disease it was 9.8 g %. Maximum haemoglobin values in ABO and Rh-D haemolytic disease were 22.6 and 17.7 g % respectively.

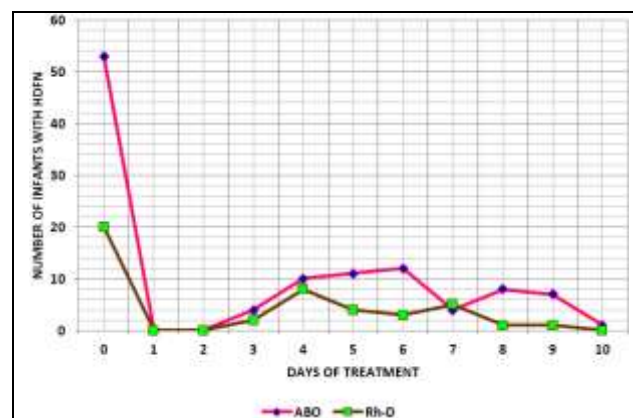


Figure 1. Duration of Treatment in Infants with ABO and Rh-D HDFN

In infants with ABO haemolytic disease 53 (48.2 %) had undergone no treatment in ICU. Remaining 51.8 % has got

3 - 10 days of treatment in neonatal ICU. In infants with Rh-D HDFN 20 (45.5 %), had undergone no treatment in ICU. Remaining 54.5 % has got 3 - 9 days of treatment in neonatal ICU.

Parameter	ABO HDFN (n = 110)	Rh-D HDFN (n = 44)	P-Value [§]
Mean duration of treatment	3.1 ± 3.3	2.9 ± 2.9	0.7044

Table 3. Comparative Analysis of Duration of Treatment in ABO and Rh-D HDFN

§ - Independent t-test has been applied as the test of significance

Mean treatment duration in infants with ABO haemolytic disease was 3.1 ± 3.3 days. Mean treatment duration in infants with Rh-D haemolytic disease was 2.9 ± 2.9 days. Median treatment duration was 3.1 and 3.6 days in ABO and Rh-D haemolytic disease respectively. The neonate was given a maximum of 10 days treatment in intensive care unit for ABO haemolytic disease. The maximum treatment duration was 9 days for Rh-D haemolytic disease. A comparison of mean values of cord blood haemoglobin of neonates in two different categories of haemolytic disease was performed. Categories included both ABO and Rh-D haemolytic disease. The resulting p value was 0.00, thus being significant. 95 % confidence interval was 1.75 to 3.50 and t value was 5.93. A comparison of mean values of duration of treatment in ICU for two different categories of haemolytic disease was done. The categories included both ABO and Rh-D HDFN. Resulting p value was 0.704 and it was an insignificant result.

DISCUSSION

HDFN has hypoxic effects of anaemia in an affected infant.¹ Augmentation of fetal erythropoiesis happens due to anaemia. This might result in extensive extramedullary haematopoiesis. This erythroblastosis is characterized by nucleated red cells in the circulation.² Most severe state of erythroblastosis and hydrops fetalis will be characterized by severe anaemia, massive oedema, hypoproteinaemia etc.³ Usual causes of anaemia in neonates are HDFN due to ABO and Rh-D antibodies.¹ This study is comparing haemoglobin levels and duration of treatment in both categories of HDFN.

In this study, comparison of mean values of cord blood haemoglobin of neonates in two different categories of haemolytic disease yielded significant. Jiri Santavy et al and Gilja et al observed that in ABO incompatible infants there will be slighter degrees of red blood cell destruction which results in slight drop in neonatal haemoglobin levels.^{5,6} Few ethnic groups may express higher number of A or B antigens in red cells. In such categories, higher levels of bilirubin along with severe anaemia with nucleated red cells in circulation was observed by Sherer et al. They also found out that even hydrops fetalis was seen in such neonates.⁷ Bjarte G et al concluded that first ABO incompatible infant was at risk for significant haemolysis. The reason behind such an observation was that the ABO antibodies were developed naturally before pregnancy itself.⁸ Mollison and Cut bush observed that the cord blood haemoglobin concentration was below normal limits in moderate to severe

ABO haemolytic disease. But it decreases in haemoglobin levels were much short lived and the anaemia was very unusual after first two weeks of life.⁹ But in a study by Rosen field and Ohno, haemoglobin concentration was slightly lower in ABO incompatible infants as compared to ABO compatible infants. Additionally they found out that haemoglobin concentration was distinctly lower in infants whose Direct Antiglobulin Test (DAT) was positive.¹⁰ In our study, even though the haemoglobin levels were decreased in ABO-HDFN group, it was significantly lesser as compared to Rh-D HDFN category.

Levine had concluded that in severe varieties of Rh-D HDFN, the cord haemoglobin concentration was significantly reduced.¹¹ Armitage had observed that before introduction of Rh immunoglobulin immunoprophylaxis for pregnant women, 50 % of affected neonates had haemoglobin levels of 14.5 g / dl or more. About 30 % had Hb values ranging from 10.5 - 14.4 g / dl and about 20 % had 3.4 - 10.4 g / dl.¹² In our study the mean was 14.470 g % and range was comparable to above mentioned study. Mollison found out that the probability of red cell survival diminished in accordance with the drop in cord haemoglobin concentration.¹³ Mollison and Cut bush found out that cord haemoglobin concentration was strongly correlated with severity of haemolytic disease.¹⁴ Crawford et al observed a minor increase in osmotic fragility in haemolytic diseased infants. Spherocytosis was found prominently in severe cases of Rh D haemolytic disease.¹⁵ Hughes jones et al concluded that anti-D levels in cord blood was not correlating with cord haemoglobin levels.¹⁶ Above mentioned findings were in par with our findings.

Last 30 to 40 years has witnessed wide and a radical change in the treatment of neonatal HDFN.⁴ Initially, by 1950s phototherapy was invented and it became the mainstay of treatment thereafter. Phototherapy improved the patient condition in HDFN significantly.⁷ Since the beginning of 1990s, several pilot studies was done on administration of IVIG as an adjuvant to the standard therapy.⁸ Two systematic meta-analyses concluded that IVIG could significantly reduce the duration of phototherapy and decrease the necessity for exchange transfusion.^{17,18} Gottstein et al opined that if the initial bilirubin levels were too high to ensue kernicterus, exchange transfusion along with intensive phototherapy, with or without IVIG, may not decrease the bilirubin levels.^{19,20} A transition from exchange transfusion and early versions of phototherapy to modern phototherapy along with high dose IVIG for serious HDFN had decreased the need for exchange transfusion.⁴ Thus, from these literature, it is very clear that different modalities of treatment and intensity of disease can affect the duration of treatment in HDFN.

Our study compared the duration of treatment in both categories of haemolytic disease and the result got was insignificant.

M.D. Eugene Kaplan et al has concluded that in Rh-D-HDFN even severely affected infants was benefitted with a trial period of phototherapy. However consideration of an exchange transfusion must be there is worsening of condition.²¹ Likewise three analyses has shown that exchange transfusion was needed, though rarely, in severe

ABO HDFN.^{22,23,24} The findings in our study was similar to these two findings.

However, Falterman CG et al has concluded that increased number of ABO antigens in red cells will result in accelerated haemolysis. This further result in jaundice, kernicterus, haemoglobinuria and even death in neonates suffering from haemolytic disease.²⁵ In a Saudi Arabian study by Miqdad AM et al, the importance of phototherapy along with IVIG was studied. They found out that IVIG when added to phototherapy in treatment of HDFN neonates, it reduced exchange transfusion rate to one-fourth.²⁶ Likewise, decrease in rate of exchange transfusion and thereby decreased hospital stay even in severe cases of HDFN might be the reason for our finding in this study.

Bowman JM opined that at birth, about 50 % of the Rh-D affected infants with DAT (Direct Antibody Test) positivity had no or mild anaemia. Thus they needed no treatment too.²⁷ Harvey Klein and David Anstee opined that in Rh D haemolytic disease, neonates might have a strong DAT positivity still having no clinical signs of disease.²⁸ But on reverse side, Bowman JM observed that one fourth of affected neonates were extremely jaundiced and developed severe disease or died.²⁷ A low reticulocyte count and thrombocytopenia was observed by Koenig in fetuses treated with intrauterine transfusion.²⁹ Bowman has added a finding that the severity of haematologic abnormalities in Rh D haemolytic disease was directly proportional to the severity of haemolysis.²⁷ Anyhow our study didn't exhibit any difference in treatment duration on both categories.

Allen FH Jr et al opined that exchange transfusion with D negative blood greatly increased the survival rate in neonates with Rh-D haemolytic disease.³⁰ Harvey Klein and David Anstee pointed out that in severe cases, intrauterine transfusion can prevent up to 80 % postnatal exchange transfusion.³¹ Sproul et al noticed that the exchange transfusion removed one fourth of bilirubin and majority of antibody coated red blood cells. However a rapid rebound of serum bilirubin level was found in many a patients necessitating further transfusion therapy.³² Sherer DM found out that erythropoiesis might be suppressed after exchange transfusion, necessitating further transfusions.⁷ Thus it is very clear that the findings related to HDFN shows a wide disparity in different studies. So to conclude with, even though our study didn't find any difference in duration of treatment for both ABO and Rh-D HDFN, a meticulous program might be mandatory for close monitoring and follow up based on case to case and region to region basis.

CONCLUSIONS

Mean haemoglobin levels in Rh-D HDFN showed a significant decrease as compared to ABO-HDFN. Duration of treatment in ABO HDFN and Rh-D HDFN presented no significant difference.

Data sharing statement provided by the authors is available with the full text of this article at jebmh.com.

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