Cholestasis in infants with immune hydrops fetalis

Şahin Takcı[1], Tuğba Alarcon-Martinez[2], Davut Bozkaya[1], Şule Yiğit[1], Ayşe Korkmaz[1], Murat Yurdakök[1]

[1]Division of Neonatology, [2]Department of Pediatrics, Hacettepe University Faculty of Medicine, Ankara, Turkey. E-mail: stakci@gmail.com

SUMMARY: Takcı Ş, Alarcon-Martinez T, Bozkaya D, Yiğit Ş, Korkmaz A, Yurdakök M. Cholestasis in infants with immune hydrops fetalis. Turk J Pediatr 2013; 55: 616-619.

Rhesus (Rh) hemolytic disease of the newborn represents a broad spectrum of symptoms in the fetus and newborn, ranging from mild to severe hemolytic anemia and hydrops fetalis. Cholestasis is a common problem in infants with immune hydrops fetalis (IHF). The aim of this study was to evaluate the incidence and course of cholestasis in infants with IHF due to Rh alloimmunization. Infants with IHF during the 10-year follow-up were retrospectively included in the study. Demographics, laboratory parameters, and prenatal and postnatal interventions were recorded. The incidence of cholestasis and certain risk factors were investigated. A total of 30 infants with IHF with a mean gestational age 33 ± 2.6 weeks were included. Of these, 15 infants (50%) survived to discharge. The incidence of cholestasis was 60% (18/30). Cholestasis was diagnosed within a median 3 (0-7) days. All cholestatic infants who survived recovered within three months. In conclusion, cholestasis in IHF is frequent, transient and has an early onset.

Key words: neonate, cholestasis, hydrops.

Rhesus (Rh) hemolytic disease of the newborn represents a broad spectrum of symptoms in the fetus and newborn, ranging from mild to severe hemolytic anemia and hydrops fetalis[1]. Anemia, excessive quantities of bilirubin, and iron overload due to hemolysis may lead to cholestasis. In addition to a few case reports associating cholestasis with Rh-hemolytic disease[2], the relationship between cholestasis and red cell alloimmunization has been documented recently by Smits-Wintjens et al.[3] However, incidence, certain risk factors and outcome of cholestasis in immune hydrops cases, which is the more severe form of Rh-hemolytic disease, have not been well described before. Therefore, the aim of this retrospective study was to evaluate the incidence and course of cholestasis in infants with immune hydrops fetalis (IHF) due to Rh alloimmunization.

Material and Methods

We performed a retrospective hospital chart review of all liveborn cases with IHF delivered at Hacettepe University Hospital (Ankara, Turkey) and admitted to the Neonatal Intensive Care Unit (NICU) during the period January 2001- June 2012. The study was approved by the Institutional Ethics Committee. IHF was described as generalized skin edema with serous effusion in one or more fetal body cavity due to Rh alloimmunization. Cholestasis was defined as conjugated bilirubin being more than 20% of total, with a minimum level of 2 mg/dl.

Infants were screened to exclude the most common causes of cholestasis in the affected children, and the following data, if available, were evaluated: serology for perinatal infections, bacterial cultures, thyroid hormones, serum alpha 1-antitrypsin levels, serum iron and ferritin levels, urine and serum amino acids, urine reducing substances, urine organic acids, other special metabolic work-up, and karyotyping and imaging studies.

Gestational age, birth weight, mode of delivery, Apgar score at the 5th minute, and prenatal and postnatal interventions, including intrauterine blood transfusions (IUTs), exchange transfusions, red cell transfusions, phototherapy durations, and drugs used for cholestasis, were recorded. Moreover, cord blood hemogram, total

and conjugated bilirubin levels, blood groups, and direct Coombs test were performed in all infants. Changes in bilirubin levels during the follow-up were also noted. The infants were excluded if conjugated bilirubin tests were not performed or their data were lost during follow-up.

Data are reported as means ± standard deviations and medians (minimum-maximum). Statistical analysis was performed using Student t test and Mann-Whitney test for continuous variables. Fisher's exact test was used for categorical variables. P values <0.05 were considered as statistically significant.

Results

During the study period, a total of 34 neonates were diagnosed as IHF due to Rh D alloimmunization. Four of them were excluded due to missing data (lack of conjugated bilirubin levels or incomplete patient file). Mean gestational age was 33±2.6 weeks and mean birth weight was 2406±649 g. Fifteen (50%) of 30 infants did not survive to discharge. Infants died at a median 2 (1-25) days. Baseline characteristics of infants with IHF are shown in Table I. Cholestasis was diagnosed in 18 infants (60%), and it was present at birth in 8 of them (26.7%). The cholestasis rate was not statistically different between surviving and non-surviving infants [11/15 (73.3%) vs 7/15 (46.7%), p>0.05]. Mean total/conjugated serum bilirubin levels were 15.4±6.4/6.9± 5.3 mg/dl in the cholestatic group and 10.5 ±7.1/0.7±0.5 mg/dl in the non-cholestatic group (p<0.05). Cholestasis was diagnosed within a median 3 (0-7) days. Comparison of the demographic, clinical and laboratory characteristics of infants with IHF in the cholestatic and non-cholestatic groups are shown in Table II. IUTs had been performed in 23 infants (range: 1-5 transfusions). The median number of IUTs in IUT-treated infants were 4 (1-5) and 1.5 (1-5) in the cholestatic and non-cholestatic groups, respectively (p>0.05).

Twelve infants received parenteral nutrition

Table I. Characteristics of Infants with Immune Hydrops Fetalis

	Infants with IHF (n=30)
Gestational age at birth, weeks[a]	33 ± 2.6
Male/female, number (%)	22/8 (73.3/26.7)
Birthweight, grams[a]	2406 ± 649
Apgar score (5th min)[b]	6 (3-8)
Hemoglobin level at birth, g/dl[a]	7.6 ± 3.5
Mortality (%)	15 (50)
Total/conjugated serum bilirubin level at birth, mg/dl[a]	4.1 ± 1.5/ 1.1± 0.8
Maximum conjugated bilirubin level, mg/dl[a]	5.5 ± 5.9
Total serum bilirubin level at the time of maximum conjugated bilirubin, mg/dl[a]	14.1 ± 6.9
Infants treated with IUT (%)	23 (76.7)
Number of IUTs in IUT-treated infants[b] (n=23)	3 (1-5)
Infants treated with ET (%)	27 (90)
Number of ETs in ET-treated infants[b] (n=27)	2 (1-6)
Number of infants with RBC transfusions (%)	22 (73.3)
Number of RBC transfusions in infants with blood transfusions[b] (n=22)	2 (1-5)
Phototherapy duration, days[b]	4 (1-8)
Number of infants who received PN (%)	12 (40)
Number of infants with sepsis (%)	4 (13.3)

ET: Exchange transfusions. IHF: Immune hydrops fetalis. IUT: Intrauterine blood transfusions. PN: Parenteral nutrition. RBC: Red blood cell.
[a]Value given as mean ± SD. [b]Value given as median (minimum-maximum).

Table II. Comparison of Characteristics in Infants with Immune Hydrops Fetalis in Cholestatic and Non-Cholestatic Groups

	Non-cholestasis group (n=12)	Cholestasis group (n=18)	P value
Gestational age at birth, weeks[a]	32.3 ± 2.8	33.5 ± 2.5	0.23
Birthweight, grams[a]	2220 ± 785	2530 ± 528	0.20
Apgar score (5th min)[b]	6 (3-7)	7 (4-8)	0.13
Hemoglobin level at birth, g/dl[a]	6.9 ± 2.8	8 ± 3.3	0.36
Mortality (%)	8 (66.7)	7 (38.9)	0.26
Total/conjugated serum bilirubin level at birth, mg/dl[a]	3.3 ± 1.4/ 0.5 ± 0.3	4.6 ± 1.5/ 1.5 ± 0.8	0.02/0.000
Maximum conjugated bilirubin level, mg/dl[a]	0.7 ± 0.6	7.6 ± 5.9	0.000
Total serum bilirubin level at the time of maximum conjugated bilirubin, mg/dl[a]	10.6 ± 7.1	15.7 ± 6.3	0.08
Number of IUTs in IUT-treated infants [b] (number of infants)	1.5 (1-5) (n=6)	4 (1-5) (n=17)	0.23
Number of ETs in ET-treated infants[b] (number of infants)	2 (1-3) (n=12)	2 (1-6) (n= 15)	0.40
Number of RBC transfusions in infants with blood transfusions[b] (number of infants)	2 (1-5) (n=12)	3 (1-4) (n=10)	0.09
Phototherapy duration, days[b]	4 (1-8)	5 (1-7)	0.29
Number of infants who received PN (%)	5 (41.7)	7 (38.9)	0.87
Number of infants with sepsis (%)	2 (16.7)	2 (11.1)	0.66

ET: Exchange transfusions. IHF: Immune hydrops fetalis. IUT: Intrauterine blood transfusions. PN: Parenteral nutrition. RBC: Red blood cell.
[a]Value given as mean ± SD. [b]Value given as median (minimum-maximum).

(PN), and of these, only four non-cholestatic infants had a prolonged course of PN (>14 days). PN was ceased or altered to intravenous dextrose solution and enteral feeding was introduced as early as possible in all cholestatic infants. The rate of cholestasis was similar between infants who did or did not receive PN [7/17 (41.2%) vs 5/13 (38.5%), p>0.05] in the cholestatic and non-cholestatic infants, respectively. Sepsis was diagnosed in four infants (*Escherichia coli* in 1, *Klebsiella pneumoniae* in 1, *methicillin-resistant Staphylococcus aureus* in 2). Cholestasis was present in two of them. All cholestatic infants who survived to discharge showed clinical and biochemical recovery from cholestasis within three months after the onset of cholestasis. Oral ursodeoxycholic acid was used in two, oral erythromycin in one and oral ursodeoxycholic acid + oral phenobarbital in one infant who survived to discharge. These four infants had high levels of conjugated hyperbilirubinemia, with levels of 12 to 19.4 mg/dl, and benefitted from the treatment.

Discussion

Prenatal management of Rh alloimmunization mainly includes anti-D immunoglobulin prophylaxis for mothers at risk and IUTs for affected fetuses. Lack of adequate management for Rh alloimmunization causes severe hemolysis, leading to fetal anemia and IHF[4]. Our hospital is a tertiary perinatal center for high-risk pregnancies, and many pregnancies with Rh alloimmunization are referred from the rural regions of Turkey. Unfortunately, some pregnancies are improperly managed and referred to us in advanced stages.

We observe cholestasis frequently in IHF cases, and it starts prenatally in a considerable number of infants in our clinical practice. The data about the association of cholestasis with IHF date to the 1960s, during which time prenatal and postnatal interventions were limited[5].

A multifactorial pathogenesis plays a role in the development of cholestasis in IHF. Inspissated bile syndrome is thought to be the major cause

of cholestasis due to excessive bilirubin load in severe hemolytic anemia[2]. Structural and functional liver damage secondary to heart failure or anemia may also play an important role in the development of cholestasis, especially in hydropic cases. Hypoxia due to anemia causes liver necrosis and induces extramedullary hematopoiesis in the liver, resulting in damage in the intrahepatic canaliculi[3-6]. Further, iron overload may contribute to cholestasis with respect to hemolysis[7].

The high incidence of cholestasis in our series can be explained by all the mechanisms mentioned herein. Although fetal bilirubin is cleared from the circulation by placental transfer, cholestasis was present in 24.1% of infants. This finding suggests that excessive bilirubin overload is present in hydropic fetuses in spite of placental elimination.

In the report of Smits-Wintjens et al.[3], there was a significant association between the number of IUTs and the risk of developing cholestasis in infants with hemolysis. It has been shown that ferritin levels were elevated in infants with Rh-hemolytic disease, and it was suggested that the high incidence of cholestasis in IUT-treated infants could be due to hepatic iron overload[3,7]. Further, the number of IUTs may reflect disease severity. Although cholestatic infants tended to have more IUTs than the non-cholestatic infants (median, 4 versus 1.5) in our study, this difference did not reach statistical significance, but this may well be due to the small sample size. The rate of exchange transfusions is quite high, as is the nature of uncontrolled hydrops cases. Exchange transfusions and duration of phototherapy did not influence the rate of cholestasis.

In our analysis, a high proportion of the cases were premature, and some of them received PN. It is well known that prematurity, prolonged PN and sepsis are strongly associated with neonatal cholestasis[8,9]. However, cholestasis occurred very early, within a median 3 days, invalidating the effect of PN in this study. Furthermore, median gestational age and the rate of sepsis were similar in cholestatic and non-cholestatic infants.

Mortality rates were high in our study population. These cases were severe and the main causes of mortality were the complications of hydrops fetalis such as prematurity, pulmonary hypoplasia, heart failure, and others. Although the difference did not reach significance, the rate of mortality in infants without cholestasis was more than that in cholestatic infants. It may be argued that in some infants who died very early, there was no time for the development of cholestasis.

In conclusion, cholestasis is very common in infants with IHF. Despite the frequency, the onset of cholestasis resolves within three months.

REFERENCES

1. Lutchman-Jones L, Wilson BD. The blood and hematopoietic system. In: Martin RJ, Fanaroff AA, Walsh ME (eds). Fanaroff and Martin's Neonatal Perinatal Medicine (9th ed). Vol. 2. St. Louis, Missouri: Elsevier; 2011: 1030-1075.
2. Allgood C, Bolisetty S. Severe conjugated hyperbilirubinaemia and neonatal haemolysis. Int J Clin Pract 2006; 60: 1513–1514.
3. Smits-Wintjens VE, Rath ME, Lindenburg IT, et al. Cholestasis in neonates with red cell alloimmune hemolytic disease: incidence, risk factors and outcome. Neonatology 2012; 101: 306-310.
4. Van Kamp IL, Klumper FJ, Bakkum RS, et al. The severity of immune fetal hydrops is predictive of fetal outcome after intrauterine treatment. Am J Obstet Gynecol 2001; 185: 668-673.
5. Dunn PM. Obstructive jaundice and haemolytic disease of the newborn. Arch Dis Child 1963; 38: 54–61.
6. Sivan Y, Merlob P, Nutman J, Reisner SH. Direct hyperbilirubinemia complicating ABO hemolytic disease of the newborn. Clin Pediatr (Phila) 1983; 22: 537–538.
7. Berger HM, Lindeman JH, van Zoeren-Grobben D, Houdkamp E, Schrijver J, Kanhai HH. Iron overload, free radical damage, and rhesus haemolytic disease. Lancet 1990; 335: 933–936.
8. Balistreri WF. Neonatal cholestasis. J Pediatr 1985; 106: 171-184.
9. Teitelbaum DH, Tracy T. Parenteral nutrition–associated cholestasis. Semin Pediatr Surg 2001; 10: 72-80.