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Frequency of Causes of Unconjugated Hyperbilirubinemia Leading to Exchange Transfusion

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ABSTRACT

Objectives: To determine the frequency of various causes of neonatal hyperbilirubinemia requiring exchange transfusion.

Methodology: This descriptive cross-sectional study was conducted from November 20, 2022, to May 19, 2023. in the Department of Pediatric Medicine at the Pakistan Institute of Medical Sciences (PIMS), Islamabad. A total of 110 neonates of either gender, requiring exchange transfusion due to unconjugated hyperbilirubinemia, were included. Inclusion criteria were neonates diagnosed with unconjugated hyperbilirubinemia based on clinical evaluation and laboratory findings. Neonates who had previously received phototherapy or had conjugated hyperbilirubinemia were excluded.

Informed consent was obtained from parents or guardians prior to enrollment. Comprehensive demographic and clinical data, including birth weight, gestational age, gender, and mode of delivery, were collected. The principal investigator assessed each neonate to identify the underlying cause of hyperbilirubinemia, which included isoimmune hemolysis, ABO incompatibility, Rh incompatibility, and other possible etiologies. Assessment involved clinical examination, review of laboratory results, and interpretation of blood group compatibility between mother and newborn. All findings were documented on a structured proforma specifically designed for the study.

Results: The mean gestational age was 37.59 ± 1.74 weeks (Table I). Of the 110 neonates, 73 (66.36%) were male and 37 (33.64%) were female, with a male-to-female ratio of 1.9:1. Among the identified causes of hyperbilirubinemia, Rh incompatibility was the most frequent, observed in 55 (55.0%) cases, followed by ABO incompatibility in 45 (40.90%) cases. Isoimmune hemolysis and other causes accounted for 5 (4.55%) cases each.

Conclusion: Rh incompatibility was found to be the most common cause of neonatal jaundice requiring exchange transfusion, followed by ABO incompatibility and isoimmune hemolysis.

Keywords: Neonatal, Jaundice, Hemolysis, Bilirubin, ABO incompatibility.

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Introduction

In the first week of life, jaundice is the most prevalent condition, affecting approximately 60% of term and 80% of preterm newborns.[1] After discharge from the hospital following birth, jaundice remains the most frequent reason for readmission.[2] Clinically, jaundice becomes evident in the skin and eyes when the total serum bilirubin (TSB) level exceeds 5–7 mg/dL. Elevated TSB in neonates is attributed to three primary factors: increased bilirubin production from red cell breakdown, reduced clearance due to immature hepatic processes, and enhanced reabsorption via enterohepatic circulation (EHC). Although hyperbilirubinemia is usually benign, in a small proportion of cases, significantly elevated serum bilirubin levels can lead to severe neurological complications.[3]

Hyperbilirubinemia is nearly universal among newborns to some degree as part of normal physiology. However, high levels of unbound, unconjugated bilirubin can cross the blood-brain barrier, resulting in neurological symptoms.[4] Hervieux first reported the association between encephalopathy and hyperbilirubinemia in 1847,[5] while Schmorl introduced the term “kernicterus” in 1903 to describe yellow staining of the basal ganglia.[6] Although some authors consider kernicterus an anatomical diagnosis and use “Bilirubin-Induced

Neurological Dysfunction" (BIND) for the clinical presentation, the terms are often used interchangeably.[7]

While most cases are physiological, pathological jaundice can occur due to conditions such as infection, liver disease, hypothyroidism, hemolysis, or metabolic disorders.[8] Bilirubin levels above 34 μmol/L (2 mg/dL) may be seen, and clinical concern arises when TSB exceeds 308 μmol/L (18 mg/dL) in otherwise healthy neonates, jaundice appears within the first 24 hours of life, levels rise rapidly, persist for more than two weeks, or the infant appears unwell. Such cases warrant further evaluation to identify the underlying cause.[9]

Kernicterus is preventable through timely intervention with phototherapy or exchange transfusion to lower unconjugated bilirubin levels to safe limits. Phototherapy effectively reduces bilirubin by converting unconjugated bilirubin into water-soluble isomers, such as lumirubin, which can be excreted in urine. However, in neonates at high risk for kernicterus, exchange transfusion (ET) remains the most rapid and effective method of reducing bilirubin levels. ET is indicated when bilirubin remains dangerously high despite intensive phototherapy and is particularly beneficial in cases of severe hemolysis.[10]

Prematurity and neonatal infections are recognized as major risk factors for neonatal jaundice[[11] Physiological jaundice involves elevated unconjugated bilirubin due to liver immaturity without other comorbidities, while pathological jaundice arises from underlying conditions that either increase bilirubin production or decrease elimination. Management of pathological jaundice necessitates treatment of the underlying cause.[12] Hemolytic disease of the fetus and newborn (HDFN) occurs at a prevalence of 1,695 per 100,000 live births. Although ABO incompatibility affects approximately 20% of births, only 1% of these cases develop clinically significant HDFN. Despite routine RhD immune globulin prophylaxis, maternal alloimmunization persists in 0.1–0.4% of at-risk pregnancies, likely due to antigens other than RhD.[13]

Given the scarcity of local data on the causes of neonatal hyperbilirubinemia requiring exchange transfusion, this study aims to identify the etiological factors in our population. The findings will contribute to baseline data and help guide preventive and therapeutic strategies in clinical practice.

Methodology

This descriptive, cross-sectional study was conducted in the Department of Pediatric Medicine at the Pakistan Institute of Medical Sciences (PIMS), Islamabad, from November 20, 2022, to May 19, 2023. CPSP Ref no. CPSP/REU/PED-2018-042-4662. A non-probability consecutive sampling technique was employed to enroll 110 neonates diagnosed with hyperbilirubinemia, irrespective of gender.

The sample size was calculated using the WHO calculator formula where prevalence of ABO incompatibility (7.6%), the margin of error (5%), and *Z* corresponds to the standard score for 95% confidence interval.

Neonates of both genders who met the operational definition of neonatal hyperbilirubinemia were included. Exclusion criteria were neonates with conjugated hyperbilirubinemia or those who had previously received phototherapy or other treatment modalities.

The study protocol was approved by the Institutional Research and Ethics Committee. Informed written consent was obtained from parents or guardians before enrollment. Demographic and clinical information, including gestational age, birth weight, gender, mode of delivery, and place of residence, was recorded on a structured proforma.

The underlying causes of hyperbilirubinemia—such as ABO incompatibility, Rh incompatibility, and isoimmune hemolysis—were identified through clinical assessment and laboratory investigations. Venous blood samples were collected and analyzed in the hospital laboratory under the supervision of a senior pathologist with over five years of post-graduate experience.

Exchange transfusion was performed using fresh whole blood (<72 hours old) compatible with both maternal and neonatal ABO and Rh blood groups. The double-volume exchange method was applied, in which small aliquots (5 mL/kg) of blood were sequentially withdrawn and replaced over 60–90 minutes. To maintain electrolyte balance, 1 mL of 10% calcium gluconate was administered for every 100 mL of blood exchanged.

Data were analyzed using SPSS version 11.5. Categorical variables (gender, gestational age category, delivery mode, preterm status, and causes of hyperbilirubinemia) were expressed as frequencies and percentages, whereas continuous variables (gestational age, total serum bilirubin levels) were presented as mean ± standard deviation (SD). Stratification was performed to control for effect modifiers such as gender, gestational age,

preterm status, place of residence, and mode of delivery. Post-stratification, the Chi-square test was applied, and a p-value <0.05 was considered statistically significant.

Results

In this study, 110 newborns with hyperbilirubinemia who received exchange transfusions were included. Seventy-nine percent of the neonates were term (≥37 weeks), with a mean gestational age of 37.59 ± 1.74 weeks. The male-to-female ratio was almost 1.9:1, with 66.36% of the patients being male, indicating a significant male predominance. While 35.45% of the newborns were born via cesarean section, the majority (64.55%) were delivered naturally through vaginal delivery. The residential distribution revealed that 53.64% of neonates were from rural areas and 46.36% were from urban settings , with 29.09% of instances involving preterm delivery (Table I).

Table I: Demographic and Clinical Characteristics of Neonates Undergoing Exchange Transfusion (n = 110)

Characteristics	Categories	N	Percentage
Gestational Age	<37 weeks	32	29.09
	≥37 weeks	78	70.91
Gender	Male	73	66.36
	Female	37	33.64
Mode of Delivery	SVD	71	64.55
	Cesarean	39	35.45
Preterm Delivery	Yes	32	29.09
	No	78	70.91
Place of Residence	Rural	59	53.64
	Urban	51	46.36

With 50.0% of cases, Rh incompatibility was the most commonly found cause of newborn hyperbilirubinemia necessitating exchange transfusion. Closely behind, ABO incompatibility was found in 40.90% of newborns. In 4.55% of instances, isoimmune hemolysis and other unidentified causes were identified (Table II). These findings show that the most common causes of severe hyperbilirubinemia in neonates that require exchange transfusions are still Rh and ABO incompatibility.

Table II: Frequency of causes of neonatal hyperbilirubinemia undergoing exchange transfusion

Causes	Frequency	Percentage
Isoimmune hemolysis	05	4.55
ABO incompatibility	45	40.90
Rh incompatibility	55	50.0
Others	05	4.55

Significant correlations between certain factors and underlying etiologies were found by stratified analysis. Rh incompatibility was more prevalent in preterm infants (p = 0.015) and was substantially correlated with gestational age (Table III). ABO incompatibility was significantly more common in female neonates (p = 0.001), while Rh incompatibility was more common in male neonates (p = 0.002), according to a gender-based comparison (Table IV). ABO and Rh incompatibility were more common in vaginally delivered babies, but no statistically significant correlations between delivery method and any particular reason were found (Table V). Furthermore, ABO incompatibility was more prevalent in newborns from rural regions (p = 0.008), whereas Rh incompatibility was significantly greater among urban dwellers (p = 0.013), according to residence-based stratification (Table VI).

Table III: Stratification of causes with respect to gestational age.

		<37 weeks (n=32)	≥37 (n=78)	P-value
Isoimmune hemolysis	Yes	03	02	0.119
	No	29	76	
ABO incompatibility	Yes	17	28	0.095
	No	15	50	
Rh incompatibility	Yes	21	34	0.015
	No	11	44	
Others	Yes	03	02	0.119
	No	29	76	

The significance of early screening for Rh and ABO incompatibilities is highlighted by these findings overall, especially in areas where these risk factors are highly prevalent. Along with improving newborn outcomes, stratified trends offer information into clinical and demographic patterns that can direct early intervention tactics.

Table V: Stratification of causes with respect to gender.

		Male (n=73)	Female (n=37)	P-value
Isoimmune hemolysis	Yes	03	02	0.758
	No	70	35	
ABO incompatibility	Yes	22	23	0.001
	No	51	14	
Rh incompatibility	Yes	44	11	0.002
	No	29	26	
Others	Yes	04	01	0.509
	No	69	36	

Table VI: Stratification of causes with respect to place of residence.

		Rural (n=59)	Urban (n=51)	P-value
Isoimmune hemolysis	Yes	04	01	0.226
	No	55	50	
ABO incompatibility	Yes	31	14	0.008
	No	28	37	
Rh incompatibility	Yes	23	32	0.013
	No	36	19	
Others	Yes	01	04	0.123
	No	58	47	

Discussion

The yellow-orange coloring of the skin and sclera caused by excessive bilirubin accumulation in tissues is a symptom of neonatal jaundice. This phenomena results from normal newborn physiology, where accelerated red cell breakdown, immature liver clearance, and improved enterohepatic circulation all contribute to elevated bilirubin levels.[14] Even though physiological jaundice goes away on its own, pathological cases that are caused by hemolysis, infection, metabolic problems, or liver dysfunction need to be evaluated right away in order to avoid kernicterus or bilirubin-induced neurological dysfunction (BIND).

According to this research, Rh incompatibility was the most common cause of unconjugated hyperbilirubinemia requiring exchange transfusion (55%), followed by ABO incompatibility (40.9%) and isoimmune hemolysis (4.6%). These findings are consistent with a number of recent studies that show hemolysis as the primary cause of exchange transfusions, especially when it occurs from blood group incompatibility. For instance, a multicenter cohort found that 63.6% of cases requiring exchange transfusion were due to hemolytic jaundice.[15] The importance of Rh and ABO incompatibility in severe neonatal hyperbilirubinemia is further highlighted by national trends.

The results of this study are consistent with Chinese genetic-clinical studies, which show a high correlation between ABO/Rh hemolysis and severe hyperbilirubinemia (odds ratio ~3.36), as well as other risk variables such weight loss and extravascular bleeding.[16] This emphasizes how crucial it is to incorporate clinical and possibly genetic markers for determining risk.

The reported rate of ABO incompatibility (~41%) is consistent with data from Iran and India, where ABO incompatibility was present in approximately 1/3 of instances of newborn jaundice. Of the 51 cases of newborn jaundice that resulted from ABO incompatibility, 24 had O-A incompatibility and 27 had O-B incompatibility. Compared to patients without ABO incompatibility, individuals with ABO incompatibility had a higher mean serum bilirubin (24.8).[17]

Improved prenatal Rh prophylaxis and increased awareness may be the cause of the relative decrease in isoimmune hemolysis when compared to previous estimations (23.1% isoimmune hemolysis vs. 4.6% in this study). A thorough evaluation supporting this trend shows that greater coverage of Rh Ig prophylaxis reduces neonatal hemolytic illness.[18]

The incidence of alloimmune hemolytic illness of the fetus and infant has declined since the invention and routine use of Rh immune globulin in Rh-negative mothers, according to a study done in the USA by Timothy M. Bahr et al., although it is still quite common in underdeveloped nations. However, one of the main causes of hemolysis is still alloimmune hemolysis brought on by different antibody/antigen pairings. Next-generation sequencing technologies and hemolysis-specific gene panels have demonstrated a high yield in identifying the etiology of hemolysis in cases when non-immune hemolysis is detected and results in hyperbilirubinemia that is prolonged or challenging to treat.[19]

A cross-sectional study was conducted by Khan et al. at Ayub Teaching Hospital in Abbottabad, where 385 full-term neonates who needed exchange transfusions (bilirubin > 19.5 mg/dL) were enrolled.[20] This demonstrates the importance of ABO/Rh incompatibility in cases of severe hyperbilirubinemia in areas that are comparable with this study.

The small sample size and single-center design of this study may have limited its generalizability. Although not investigated, genetic predispositions such UGT1A1 variations may affect the severity of the condition. To enhance risk stratification, future studies should include genetic testing in addition to clinical assessment.

Conclusion

This study has shown that the Rh incompatibility is the most common cause for neonatal jaundice followed by ABO incompatibility and iso immune hemolysis. So, we recommend that early identification and management of these factors should be done in order to decrease morbidity and mortality by this deadly but preventable condition.

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