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Original Article / Orijinal Araştırma

Role of COHb Level in Newborns with ABO Blood Group Incompatibility in Predicting Newborn Jaundice Risk

ABO Kan Grubu Uyuşmazlığı Olan Yenidoğanlarda Kord Kan COHb Düzeyinin Yenidoğan Sarılığı Riskini Öngörmedeki Rolü

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ABSTRACT

Objective: Neonates with hyperbilirubinemia are at risk of brain damage, and at least two-thirds of neonates show clinical signs of jaundice in the first week of life. To predict the correlation between cord carboxyhemoglobin (COHb) with postnatal 24th-hour total serum bilirubin (TSB) level in newborns with ABO incompatibility.

Methods: This retrospective cohort study included newborns older than 35 weeks of gestation who were followed up in the Neonatal Intensive Care Unit of Tokat Gaziosmanpaşa University Hospital between January 2019 and December 2023. Patients were divided into three groups; Group 1: ABO incompatibility with direct Coombs (DC) positive newborns, Group 2: ABO incompatibility and DC negative newborns, and Group 3: ABO incompatibility with no known hemolysis risk factors.

Results: A total of 292 patients in 3 groups were included in the study. Group 1 consisted of 93 patients, Group 2 consisted of 99, and Group 3 consisted of 100. The mean newborn cord COHb was 1.59±0.56%, the mean cord bilirubin was 3.12±2.05 mg/dL, the mean 24-h TSB was 6.40±1.99 mg/dL, and the mean cord blood gas hemoglobin was 18.06±2.57 g/dL. In the first group, the correlation between cord COHb and 24-h TSB was high and statistically significant. In the second group, the correlation between cord COHb and 24-h PTH was low and statistically significant.

Conclusion: The use of COHb as a predictor of 24-h postnatal TSB levels in ABO-incompatible neonates is plausible.

Keywords: Neonatal jaundice, COHb, ABO incompatibility, Kernicterus

ÖZ

Amaç: Hiperbilirubinemili yenidoğanlarda potansiyel beyin hasarı riski vardır ve yenidoğanların en az üçte ikisi yaşamlarının ilk haftasında klinik sarılık belirtileri gösterir. ABO uyuşmazlığı olan yenidoğanlarda kord karboksihemoglobin (COHb) ile postnatal 24. saat total serum bilirubin (TSB) düzeyi arasındaki ilişkiyi öngörmeyi hedefledik.

Yöntem: Retrospektif kohort analizi kullanılarak yapılan bu çalışma, Ocak 2019 ile Aralık 2023 tarihleri arasında Tokat Gaziosmanpaşa Üniversite Hastanesi Yenidoğan Yoğun Bakım Ünitesi'nde takip edilmiş 35. gebelik haftasından büyük yenidoğanlar ile gerçekleştirildi. Hastalar üç gruba ayrıldı; Grup 1: ABO uyuşmazlığı ve direkt Coombs (DC) pozitif yenidoğanlar, Grup 2: ABO uyuşmazlığı ve DC negatif yenidoğanlar, Grup 3: ABO uyuşmazlığı olmayan ve bilinen hemoliz risk faktörü olmayan yenidoğanlar.

Bulgular: Çalışmaya 3 grupta toplam 292 hasta dahil edildi. Grup 1, 93 hastadan, Grup 2, 99 hastadan ve Grup 3, 100 hastadan oluşuyordu. Yenidoğan kord COHb ortalaması %1,59±0,56, kord bilirubini ortalaması 3,12±2,05 mg/dL, 24. saat TSB ortalaması 6,40±1,99 mg/dL, kord kan gazındaki hemoglobin ortalaması 18,06±2,57 g/dL olarak belirlendi. Birinci grupta kord COHb ile 24. saat TSB arasındaki korelasyon yüksek ve istatistiksel olarak anlamlıydı. İkinci grupta kord COHb ve 24. saat TSB korelasyonu düşük ve istatistiksel olarak anlamlıydı.

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Sonuç: Bu çalışma, ABO uyuşmazlığı olan yenidoğanlarda doğum sonrası 24. TSB düzeyini tahmin etmede COHb kullanımının akla yatkın olduğu sonucunu ortaya koymaktadır.

Anahtar Kelimeler: Yenidoğan sarılığı, COHb, ABO uyuşmazlığı, Kernikterus

INTRODUCTION

Neonatal jaundice is frequently noted within the first 2 weeks of life and is the most common reason for hospital admission of neonates. As such, it is a major issue for clinicians and families.[1,2] Approximately 5-10% of neonatal jaundice is pathological and requires treatment.[3] Neonatal jaundice is generally a consequence of unconjugated hyperbilirubinemia, a condition that results from a limited ability to conjugate the increased bilirubin levels that occur in the first days of life. Severe cases may lead to serious neurological complications, kernicterus, and even death.[4,5]

The potential risk of bilirubin-induced brain damage underscores the critical importance of early detection of neonatal jaundice.[6,7] Identification of infants at risk of severe hyperbilirubinemia and early diagnosis are critical and can reduce the need for rehospitalization and prevent possible complications.[8,9]

For social, economic, and medical reasons, the duration of post-birth hospitalization among newborns and their mothers has reduced, and early discharge rates have increased. Because this increases the risk of severe hyperbilirubinemia and its complications, various methods are used to predict each infant's risk of neonatal jaundice prior to hospital discharge.[10]

ABO blood group incompatibility is an important risk factor for neonatal jaundice.[11] Bilirubin is a byproduct of heme molecule metabolism. However, in newborns with ABO blood group incompatibility, there is an enzymatic reaction between heme molecules, heme oxygenase, and iron during the breakdown of hemoglobin that causes the release of carbon monoxide (CO) and bilirubin into the body.[12] This catabolism indicates that measurement of carboxyhemoglobin (COHb) levels in cord blood may be a useful tool for detecting risk and early diagnosis of hyperbilirubinemia in newborns.[13]

This study aimed to investigate the correlations between cord blood gas COHb and total serum bilirubin (TSB) levels and ABO blood group incompatibility in newborns at postnatal hour 24 and the neonatal jaundice risk prediction accuracy.

METHODS

This retrospective cohort study included neonates born after >35 weeks of gestation who were hospitalized in the Neonatal Intensive Care Unit at Tokat Gaziosmanpaşa University Hospital between January 2019 and December 2023. The study was conducted with the permission of Tokat Gaziosmanpaşa University Hospital Ethics Committee (date: 18.01.2024, decision no: 83116987-039). Data were collected from patient medical records. Three patient groups were analyzed. Group 1 comprised patients with ABO blood group incompatibility between mothers and infants and positive direct Coombs (DC) results in newborns. Group 2 comprised patients with ABO blood group incompatibility and DC-negative newborns. Group 3 comprised patients without known ABO blood group incompatibility and hemolysis risk factors.

Exclusion criteria:

1. Maternal thyroid function test abnormalities;
2. Inadequate oral intake;
3. Congenital anomalies;
4. Sepsis or suspected sepsis;
5. Rh incompatibility;
6. Glucose-6-phosphate deficiency;
7. Incomplete patient data;
8. Hospitalization beyond postnatal hour 24;
9. Metabolic disorder.

Inclusion criteria:

1. Delivery after 35 week gestation;
2. ABO-incompatible neonates with or without positive results on a DC test;
3. Newborns without possible jaundice, such as those with transient respiratory tachypnea and congenital pneumonia, were included as a control group;
4. Delivery took place in our hospital;
5. TSB measurement 24 hours after birth and cord blood gas analysis at birth;
6. Complete patient data.

Only patients with blood ABO blood group incompatibility were included. Patients with other causes of hemolysis were excluded to reduce possible variables affecting outcomes. The use of phototherapy was based on the guidelines of the Turkish Neonatal Society and the American Academy of Pediatrics.[14,15]

Demographic, clinical, and pathological information included sex, gestational age, birth weight, cord blood gas values (COHb, Hb, hematocrit, and bilirubin), TSB measured at the 24th hour, DC test result, mode of delivery, and whether or not phototherapy was administered.

Among patients whose TSB levels identified them as having a medium-high risk of jaundice, we determined whether they had received phototherapy based on the record of their daily progress reports.

Blood bilirubin was measured using an ABL 800 Flex (Danaher Corp., Washington DC, USA) (195 µL). TSB was measured using a Roche Diagnostics COBAS six thousand AutoAnalyzer (Roche Diagnostics, Indianapolis, IN, USA).

Statistical Analysis

Quantitative variables were presented as mean and standard deviation and qualitative variables as number (n) and percentage (%). Differences in quantitative measurements between independent groups were determined using one-way analysis of variance (ANOVA). Qualitative values were analyzed using chi-square tests. Tukey's honestly significant difference test and Tamhane's T2 test were used for multiple comparisons. Pearson's correlation coefficient was used to identify relationships between quantitative variables. Receiver operating characteristics (ROC) curves were applied to determine the cut-off values of the variables by group; the area under the curve (AUC) was also evaluated. P values <0.05 were considered statistically significant. All analyses were performed using Statistical Package for Social Sciences (SPSS) v.22. statistical software (SPSS Inc., IBM Corp., Armonk, NY, USA).

RESULTS

A total of 292 patients were included in the study. Group 1 (ABO incompatibility and positive DC) comprised 93 patients, Group 2 (ABO incompatibility and negative DC) comprised 99 patients, and group 3 (no known risk factor for blood incompatibility) comprised 100 patients.

Of the 292 patients, 161 (55.8%) were male and 129 (44.2%) were female. There were 199 (68.2%) patients with negative neonatal DC test results, 29 (9.9%) with 1+ results, 42 (14.4%) with 2+ results, 21 (7.2%) with 3+ results, and 1 (0.3%) with a score of 4+ on the DC test. Among the deliveries, 255 (87.3%) were cesarean section (C/S) and 37 (12.7%) were normal vaginal deliveries. Among the included patients, 174 (59.6%) received phototherapy while 118 (40.4%) did not (Table 1).

The mean gestational week at birth was 37.27 (±1.63) and the mean birth weight was 3036.65 (±548.12) g. The mean newborn cord COHb level was 1.59 (±0.56)%, the mean cord bilirubin level was 3.12 (±2.05) mg/dL, and the mean 24-hour TSB level was 6.40 (±1.99) mg/dL. The mean cord blood gas concentration (Hb) was 18.06 (±2.57) g/dL, and the mean cord heart rate (HTH) was 54.57 (±7.95)% (Table 2).

In Group 1, there was a strong significant correlation between cord COHb and 24-h TSB (p<0.001*, r=0.603). In Group 2, there was a weak but significant correlation between cord COHb and 24-h TSB (p<0.001*, r=0.378). In Group 3, there was no correlation between cord COHb and 24-h TSB (p=0.675, r=0.042).

ROC analysis of cord COHb and 24-h TSB in Group 1 revealed a significant relationship (p<0.001). The threshold

Table 1. Distributions of qualitative values (n=292)

		n	%
Group	1. Group	93	31.8
	2. Group	99	33.9
	3. Group	100	34.2
Sex	Male	163	55.8
	Female	129	44.2
Neonatal direct Coombs	Negative	199	68.2
	1+	29	9.9
	2+	42	14.4
	3+	21	7.2
	4+	1	0.3
Mode of delivery	C/S	255	87.3
	NVD	37	12.7
Phototherapy	No	118	40.4
	Yes	174	59.6

N: Number, %: Percent, +: Positive.
C/S: Cesarean section, NVD: Normal vaginal delivery

Table 2. Distributions of quantitative measures (n=292)

	Average	Standard deviation	Minimum	Maximum
Gestational week	37.27	1.63	33.00	42.00
Birth weight (grams)	3036.65	548.12	2000.00	5000.00
Cord COHb (%)	1.59	0.56	0.10	3.70
Cord bilirubin (mg/dL)	3.12	2.05	0	10.40
24th hour TSB (mg/dL)	6.40	1.99	1.00	11.80
Cord HB (gr/dL)	18.06	2.57	10.10	25.30
Cord HTC (%)	54.57	7.95	30.30	77.10

N: Number, %: Percent, Cord HB: Cord hemoglobin, Cord HTC: Cord hematocrit, mg/dL: Milligram/deciliter, TSB: Total serum bilirubin, COHb: Carboxyhemoglobin

value of cord COHb was 1.8%. The specificity and sensitivity were 100% and 54.88, respectively (Figure 1). There was also a significant relationship between these two variables in Group 2 ($p<0.009$). In this group, the threshold cord COHb level was 1.7%. The specificity and sensitivity were 80.39% and 45.83%, respectively (Figure 2).

The relationship between cord COHb level and receipt of phototherapy treatment was analyzed in Group 1, and a moderately significant correlation was found ($p<0.001$, $r=0.568$).

The number of patients in groups 1, 2, and 3 whose cord blood gas bilirubin and subsequent TSB levels exceeded 5 mg/dL were 18 (19%), 14 (14%), and 6 (6%) patients, respectively.

DISCUSSION

We found a significant correlation between cord COHb levels and 24-h TSB levels in newborns with ABO incompatibility. This correlation was significant regardless of whether the patient had a positive DC test result although it was stronger in those with positive DC results.

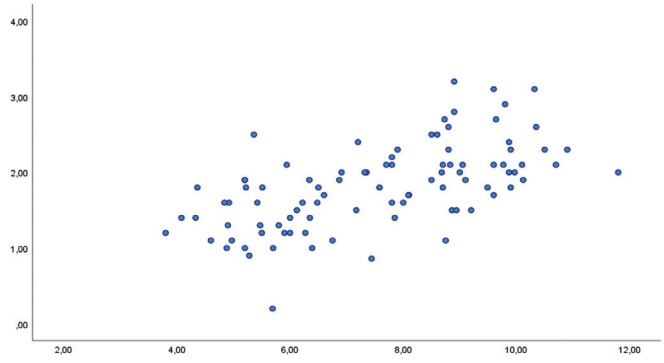

Figure 1. The scatter plot shows the relationship between the cord capillary tube COHb level (vertical axis) and 24th-hour TSB (horizontal axis) in Group 1

TSB: Total serum bilirubin, COHb: Carboxyhemoglobin

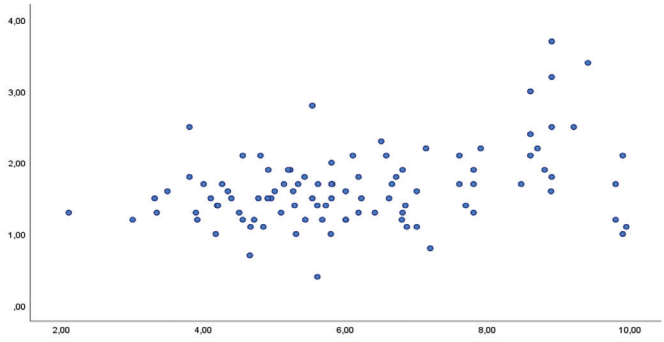

Figure 2. The scatter plot shows the relationship between the cord capillary tube COHb level (vertical axis) and 24th-hour TSB (horizontal axis) in Group 2

TSB: Total serum bilirubin, COHb: Carboxyhemoglobin

A 2021 study by Tıraş et al.[16] divided patients into the same three groups as in the present study to determine whether cord blood COHb levels can be used as a predictor of severe hyperbilirubinemia in jaundiced term neonates with and without positive DC test results. However, the authors did not find COHb levels to be a better predictor than the DC test of severe hyperbilirubinemia in term infants. The differing results between this study and ours may be due to the higher number of patients in the present study.

There are numerous national and international recommendations and guidelines and a plethora of studies on the early detection and monitoring of hyperbilirubinemia.[17] For example, the approach, follow-up, and treatment guidelines for neonatal jaundice were revised by the Turkish Neonatology Association in 2023. Hyperbilirubinemia can cause severe and irreversible brain damage in newborns. To define physiological-pathological jaundice, it is important to consider the gestational and postnatal ages of the infant, assess the risks, and evaluate TSB hourly using the bilirubin nomogram. Although progress has been made in neonatal health in recent years, some health system-related problems remain. These include high C/S rates, early discharge after birth (less than 24 hours), inadequate checkup policies for 48 hours after discharge, and checks performed by family physicians with insufficient knowledge of neonatal health.[14] For these reasons, we aimed to analyze the use of cord COHb as a potential risk predictor of neonatal jaundice.

TSB levels are currently the gold standard for diagnosing neonatal jaundice. According to the bilirubin nomogram, bilirubin values should be interpreted according to the patient's age in hours. This nomogram allows us to monitor changes in bilirubin values over time and predict the development of hyperbilirubinemia. However, adequate samples are not always obtained, and the procedure can be uncomfortable or painful for patients. An alternative method for measuring bilirubin levels at the bedside is the capillary tube method, which requires only a small blood sample. This method is less painful and more accessible. Bilirubin can also be measured on the skin surface using transcutaneous bilirubin measurement, eliminating the need for a blood sample. The ubiquitous use of this method decreased the incidence of severe hyperbilirubinemia and rehospitalization for phototherapy treatment. However, this method is unreliable for newborns receiving phototherapy and for those with darker skin.[18]

ABO blood group incompatibility is one of many conditions that can cause neonatal hyperbilirubinemia. COHb and

bilirubin, which are produced as a result of the catabolism of hemoglobin, can be measured concurrently by blood gas analysis. A recent study investigated the relationship between cord COHb levels and severe hyperbilirubinemia. The study found that cord COHb can be used for the early diagnosis and treatment of hemolysis. When the cord COHb cut-off value was set at 2.2%, the authors found a sensitivity of 80.8% and a specificity of 95.5% in its ability to predict severe hyperbilirubinemia.[19] In our study, a cord COHb cut-off value of 1.8% in patients with ABO blood incompatibility who were also DC-positive produced a specificity of 100% and sensitivity of 54.88%. A cord COHb cut-off value of 1.7% in patients with ABO blood incompatibility that were also DC-negative produced a sensitivity of 80.39% and specificity of 45.83%. In a similar study, the correlation between cord COHb levels and TSB levels during follow-up in patients with ABO blood group incompatibility and DC test positivity was found to be significantly higher compared with the control group.[11] The findings of this study are consistent with the present findings.

Lozar-Krivec et al.[20] found that newborns with ABO blood group incompatibility who were later diagnosed with hyperbilirubinemia had significantly higher cord COHb values than those without ABO blood group incompatibility. In the same study, a cord COHb cut-off value of 1.7% for confirming hemolysis in ABO alloimmunization of 1.7% resulted in 72% sensitivity and 97% specificity.[20] This cord COHb cut-off value was close to our own but had a pronounced difference in sensitivity and specificity. While sensitivity was higher in our study, specificity was significantly higher in their study.

Guney Varal et al.[21] found that newborns who received phototherapy treatment upon admission had significantly higher cord COHb levels than those who did not receive treatment. This trend continued in the following hours. They also found a positive correlation between the first and subsequent cord COHb levels and TSB and that direct antiglobulin test positivity significantly affected the need for phototherapy. They showed that cord COHb levels ≥0.95% had a sensitivity of 90% and a specificity of 88% in terms of the need for phototherapy (TSB ≥95% percentile). Although the cord COHb cut-off value was much lower than that of our study, they achieved high sensitivity and specificity.

Blood CO can be measured invasively from cord blood gas, and end-tidal CO corrected for inhaled CO concentration (ETCOc) can be measured noninvasively. In our study, COHb levels were measured using an invasive method. We compared our study data with that of previous research using the ETCOc method. In a study by Christensen et al.[22] based on ETCOc measurements, 11 of 100 patients had ABO blood incompatibility. None of the 100 were rehospitalized for jaundice treatment compared with 2.99 rehospitalizations per 100 control neonate who had TSB values >75th percentile. Bhutani et al.[23] measured ETCOc and TSB in 641 newborns 30 (±6) hours before discharge and in patients with ETCOc ≥1.7 ppm. TSB was significantly higher. These two studies using ETCOc had different COHb results from one another, with Christensen et al.[22] finding results that differed from our own and Bhutani et al.[23] producing similar outcomes to ours.

Maisels and Kring[24] the relationship between TSB and ETCO levels between the first and fifth days of life in patients diagnosed with neonatal jaundice and 164 healthy newborns. A positive correlation was found for ETCO based on the 75th percentile of TSB, and a statistically significant difference was observed between the two groups. The study also found a positive relationship between newborn COHb levels measured by ETCO and TSB levels.

Bhatia et al.[25] found that hemolysis was more commonly observed in babies with an ETCOc ≥1.8 ppm than those with an ETCOc <1.8 ppm. This was linked to higher TSB levels, a rapid increase in TSB, and longer phototherapy duration. ETCOc values can be used as an indicator of hemolysis and a predictor of significant hyperbilirubinemia development in newborns. Although the CO measurement method was different, our results suggest that COHb and ETCOc measurements are similarly effective in predicting the risk of neonatal jaundice.

Study Limitations

Our study has some limitations. Newborns born at less than 35 weeks' gestation were excluded. Only hemolysis patients with ABO blood group incompatibility were examined, and the data were gathered retrospectively. A more comprehensive prospective study is needed to verify our findings.

CONCLUSION

We found the cord blood gas COHb level to be a meaningful parameter for predicting the TSB level within the first 24 hours after birth in DC-positive and DC-negative patients with ABO incompatibility. The accuracy of hyperbilirubinemia prediction using COHb levels needs to be verified in further research, which should investigate causes beyond ABO incompatibility.

Ethics

Ethics Committee Approval: The study was conducted with the permission of Tokat Gaziosmanpaşa University Hospital Ethics Committee (date: 18.01.2024, decision no: 83116987-039).

Informed Consent: Because the study was designed retrospectively, no written informed consent forms were obtained from the patients.

Authorship Contributions

Surgical and Medical Practices: K.D., Ş.T., Concept: K.D., Ş.T., Design: K.D., Ş.T., Data Collection or Processing: K.D., Ş.T., Analysis or Interpretation: K.D., Ş.T., Literature Search: K.D., Ş.T., Writing: K.D.

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