



Correlation of Capillary Tube and Transcutaneous Methods with Serum Biochemistry for Bilirubin Levels in Neonates with Indirect Hyperbilirubinemia

İndirekt Hiperbilirubinemili Yenidoğanlarda Bilirubin Düzeyleri için Kapiller Tüp ve Transkütan Yöntemlerinin Serum Biyokimyası ile Korelasyonu

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Abstract

Aim: This study aimed to compare transcutaneous bilirubin (TcB) measurement, which is a noninvasive, simple, fast, and inexpensive method for treating patients with indirect hyperbilirubinemia (IHB), to total serum bilirubin (TSB) measurement values, which are the standard method. In addition, the study included contemporaneous capillary tube (CT) bilirubin values to determine the relationship. We aimed to investigate the relationship between TSB and TcB levels before and after treatment for IHB in patients.

Material and Method: Between January 1 and July 1, 2023, newborns aged 37 to 42 weeks admitted to the neonatal intensive care unit at the Faculty of Medicine, Gaziosmanpaşa University (Tokat, Turkey) with IHB were included. Patients who received phototherapy for IHB were evaluated retrospectively. TcB measurements were performed in patch-covered ears shortly before and 24 h after treatment (together with TSB and CT measurements). The data of patients who underwent phototherapy for IHB and had their TcB, TSB, and capillary tube bilirubin levels assessed before and after treatment were analyzed retrospectively. Measurements were taken at the beginning and 24 h of treatment.

Results: Seventy-two patients were term neonates. Six patients were excluded from the study because they did not meet the criteria. The mean gestational age was 37.23 ± 0.60 weeks (37-40 weeks), and the mean weight was $2,751.52 \pm 432.84$ g (2,100-4,550 g). Twenty-seven (40.9%) of the newborns were male. The direct Coombs test resulted in 1+ in five newborns and 4+ in one newborn. There was a statistically significant difference between TcB bilirubin levels and TSB measurement values before and after treatment. Furthermore, there was a statistically significant difference between the CT bilirubin and TSB measurement values before and after treatment.

Conclusion: Our study demonstrated a statistically significant difference in TSB bilirubin values using the TcB and CT methods before and after treatment. Although TcB and CT methods can be used in IHB screening, we do not recommend using them instead of the TSB measurement method for treatment and follow-up.

Keywords: Bilirubin, newborn, kernicterus, transcutaneous

Öz

Amaç: Çalışmamızda indirekt hiperbilirubinemili (İHB) hastaların tedavisinde noninvasiv, basit, hızlı ve ucuz bir yöntem olan transkütan bilirubin (TcB) ölçümü ile standart yöntem olan total serum bilirubin (TSB) ölçüm değerlerinin karşılaştırılması amaçlanmıştır. Ayrıca, çalışmaya eş zamanlı çalışılmış olan kapiller tüp bilirubin değerleri de dahil edilmiştir. Hastalarda İHB tedavisi öncesi ve sonrası TSB ve TcB düzeyleri arasındaki ilişkiyi araştırmayı amaçladık.

Gereç ve Yöntem: 1 Ocak - 1 Temmuz 2023 tarihleri arasında Gaziosmanpaşa Üniversitesi Tıp Fakültesi (Tokat, Türkiye) Yenidoğan Yoğun Bakım Ünitesinde İHB nedeniyle hastane yatışı yapılmış olan 37-42 haftalık hastalar çalışmaya dahil edildi. İHB için fototerapi alan hastalar retrospektif olarak değerlendirildi. TcB ölçümleri tedaviden, hemen önce ve 24 saat sonra (TSB ve kapiller tüp ölçümleri ile birlikte) patch ile kapalı kulaklardan yapıldı. İHB nedeniyle fototerapi uygulanan ve tedavi öncesi-sonrasında TcB, TSB ve kapiller tüp bilirubin düzeyleri değerlendirilen hastaların verileri retrospektif olarak analiz edilmiştir. Ölçümler tedavinin hemen başlangıcında ve 24. saatinde eş zamanlı yapılmıştır.

Bulgular: Yetmiş iki hasta term yenidoğandı. Altı hasta kriterleri karşılamadığı için çalışma dışı bırakıldı. Ortalama gebelik yaşı 37.23 ± 0.60 hafta (37-40 hafta) ve ortalama ağırlık $2,751.52 \pm 432.84$ g (2,100-4,550 g) idi. Yenidoğanların yirmi yedisi (%40,9) erkekti. Direkt Coombs testi beş yenidoğanda 1+ ve bir yenidoğanda 4+ olarak sonuçlanmıştır. Tedavi öncesi ve sonrası TcB bilirubin düzeyleri ve TSB ölçüm değerleri arasında istatistiksel olarak anlamlı bir fark vardı. Ayrıca, tedavi öncesi ve sonrası kapiller tüp bilirubin ve TSB ölçüm değerleri arasında istatistiksel olarak anlamlı bir fark vardı.

Sonuç: Çalışmamızda tedavi öncesi ve sonrası TcB ve kapiller tüp yöntemleri kullanılarak TSB bilirubin değerlerinde istatistiksel olarak anlamlı bir fark olduğu gösterilmiştir. TcB ve kapiller tüp yöntemleri İHB taramasında kullanılabilir de tedavi ve takip kararı için TSB ölçüm yöntemi yerine kullanılmasını önermiyoruz.

Keywords: Bilirubin, yenidoğan, kernicterus, transkütan



INTRODUCTION

Indirect hyperbilirubinemia (IHB) is a common disorder in newborns^[1] and is estimated to occur in 60% of term and 80% of preterm newborns,^[2] which is a physiological condition that often occurs in the postnatal period. It is not a major disease but a physical finding due to many causes. Severe IHB is considered pathologic. IHB is characterized by the accumulation of yellow-orange pigment bilirubin in the skin, sclera, and other tissues. Because IHB is not a specific disease and its causes are multiple different disorders, preventive and therapeutic approaches to IHB or hyperbilirubinemia are typically nonspecific.^[3] All approaches to IHB aim to prevent kernicterus, which can occur due to increased unconjugated bilirubin levels. Kernicterus is a serious condition that may result in permanent damage. If kernicterus occurs, cerebral palsy, bilateral sensorineural hearing loss, and often upward gaze limitation are typically observed.^[4] The 2004 American Academy of Pediatrics Hyperbilirubinemia Subcommittee guidelines recommend the total serum bilirubin (TSB) or transcutaneous bilirubin (TcB) measurement before hospital discharge to assess the risk of severe hyperbilirubinemia in all newborns.^[5]

TSB measurement remains the gold standard method in IHB, but it is invasive and painful because it is performed by venous or heel blood sampling.^[6]

Furthermore, despite the low risks associated with blood collection, the risks of complications such as iatrogenic anemia, puncture site infection, bacteremia, and osteomyelitis cannot be ignored.^[7] TcB measurements estimate the total serum bilirubin levels using multi-wavelength spectral reflections from the skin surface.^[8] Several studies have reported that the use of TcB or TSB measurements during IHB screening reduces the incidence of neonates with severe hyperbilirubinemia, the hospitalization rate for phototherapy, and the number of infants receiving phototherapy.^[9-11] However, the accuracy of TcB measurements during phototherapy is lower than TSB. Therefore, TcB measurement is unreliable in neonates receiving phototherapy.^[12-14] Differences may be seen between TcB measuring devices. If TcB is used instead of TSB measurement in the clinic, TcB values should be compared with TSB values, and their accuracy should be tested.^[15]

For that reason, our study aimed to compare TcB measurement (a noninvasive, easy, fast, and inexpensive method) and TSB measurement values (the standard method for treating IHB). At the same time, CT bilirubin values were also included in the study for correlation purposes. We examined the correlation of TcB measurement with TSB values before and after treatment in patients followed up with a diagnosis of IHB.

MATERIAL AND METHOD

The study was carried out with the permission of Tokat Gaziosmanpasa University Ethics Committee (Date: 18.01.2024, Decision No: 83116987-038). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Newborns with gestational ages of 37-42 weeks who were hospitalized with IHB were enrolled between January 1, 2023, and July 1, 2023, in the neonatal intensive care unit of the Gaziosmanpasa University School of Medicine (Tokat, Türkiye). Patients with confirmed IHB were retrospectively investigated. Only term newborns (37-42 weeks) were included in the study. Newborns with congenital anomalies, applied exchange transfusion, previous phototherapy treatment, ABO-Rh blood group incompatibility, and pre/post-maturity were excluded from the study. Patients who received phototherapy for the first time and had no transfusion history were included in the study. In patients treated for IHB, TcB measurements were performed in patch-covered ears just before and 24th hours after treatment (at the same time as TSB and CT measurements). The data of patients who received phototherapy for IHB and whose TcB, TSB, and capillary tube bilirubin levels were measured before and after treatment were examined retrospectively. Six patients who did not meet the study criteria (due to a positive direct Coombs test) were excluded from the study. Bilirubin measurements were obtained simultaneously from routine data at the 0th and the 24th hours after the start of IHB treatment. TcB was determined by Bilicare (Gerium 2016, Israel) TcB-measuring device. Measurements from the right and left ears of each patient at pre- and post-treatment were analyzed and obtained from hospital records. The arithmetic mean of measurements taken from the right and left ears was combined into a single TcB value. Those with CT and TSB values were evaluated simultaneously. TcB values were also measured at the same time. The CT bilirubin values were measured using the ABL 800 Flex (195 µL, Danaver, 2200 Pennsylvania Avenue, NW). TSB values were determined using Roche Diagnostics adapted to COBAS 6000 AutoAnalyzer (Roche Diagnostics, Indianapolis, IN).

Statistical Analysis

Descriptive statistics were used to provide information about the general characteristics of the study groups. Data for quantitative variables are means and standard deviations ($x \pm SD$). Data for qualitative variables were defined using number (n) and percentage (%). One Sample T Test was used to examine whether the difference between variables differed from zero. P values were considered statistically significant when calculated to be less than 0.05. Ready-made statistical software was used in the calculations. (IBM SPSS Statistics 22, SPSS Inc., an IBM Co., Somers, NY, USA).

RESULTS

All 72 patients were term neonates. Sixty-six patients met the criteria. The mean gestational age was 37.23 ± 0.60 weeks (37-40 weeks), and the mean weight was 2751.52 ± 432.84 g (2100-4550 g). Twenty-seven (40.9%) neonates were male, and thirty-nine (59.1%) neonates were female. The direct Coombs (DC) test was also detected as 1+ in five neonates and 4+ in one neonate. These six patients (with positive DC

who did not meet the study criteria were excluded from the study. All patients were delivered by cesarean section. 32 (31.9%) patients had 0 Rh+, 5 (6.9%) had B Rh, 24 (34.7%) had 0 Rh+, and 5 (6.9%) patients had AB Rh+.

A statistically significant difference was observed between pre- and post-treatment TcB and TSB values ($p < 0.001$; **Tables 1 and 2**).

Table 1. Accuracy of pretreatment TcB values compared to serum bilirubin values.

	Mean	Standard Deviation
TcB (mg/dL)	14.49	3.11
TSB (mg/dL)	13.67	2.90
Difference	0.8201	1.8787
95% Confidence Interval	(-2.86215) – (+4.502352)	
p	<0,001*	

N: Sixty-six (66), TcB: Transcutaneous bilirubin, TSB: Total serum bilirubin

Table 2. Accuracy of post-treatment TcB values compared to serum bilirubin values.

	Mean	Standard Deviation
Post-Treatment TcB (mg/dL)	11.11	3.10
Post-Treatment TSB (mg/dL)	10.64	2.80
Difference	0.4743	1.98391
95% Confidence Interval	(-3.41416) – (+4.362764)	
p	0,046*	

N: Sixty-six (66), TcB: Transcutaneous bilirubin, TSB: Total serum bilirubin

A statistically significant difference was observed between CT and TSB values pre- and post-treatment (**Table 3 and 4**).

Table 3. Accuracy of pretreatment capillary tube bilirubin values compared to serum bilirubin values.

	Mean	Standard Deviation
CT Bilirubin (mg/dL)	14.89	3.25
TSB (mg/dL)	13.67	2.90
Difference	1.22	1.57
95% Confidence Interval	(-1.86311) – (+4.301912)	
p	<0,001*	

N: Sixty-six (66), CT: Capillary tube, TSB: Total serum bilirubin

Table 4. Accuracy of post-treatment capillary tube bilirubin values compared to serum bilirubin values

	Mean	Standard Deviation
CT Bilirubin(mg/dL)	11.34	3.08
TSB(mg/dL)	10.64	2.80
Difference	0.71	1.35
95% Confidence Interval	(-1.94122) – (+3.352423)	
p	<0,001*	

N: Sixty-six (66), CT: Capillary tube, TSB: Total serum bilirubin

DISCUSSION

Our study analyzed the data of 66 newborn patients who underwent IHB treatment and had their measurements measured at the 0th and 24th hours. TSB was accepted as the gold standard and compared with TcB and CT method measurements. In our study, there was a statistically significant difference between pretreatment and post-

treatment TSB measurements and TcB and CT. Our findings do not support using the TcB and CT bilirubin methods instead of the TSB method to determine treatment and follow-up, although they are used as pretreatment screening methods for IHB. Because there is no subgroup information on blood group incompatibility in the patients in our study, subgroup testing may yield different results.

IHB is a major disorder occurring in term newborns on the first day of life.^[16] In the first week of life, 60-70% of term newborns are diagnosed with IHB, and an average of 85% of newborns admitted to the hospital in the first week of life are neonates diagnosed with IHB.^[22] IHB is usually harmless and self-limiting. However, kernicterus may cause permanent damage to brain tissues due to very high bilirubin levels. Therefore, the diagnosis and treatment of IHB are critical.^[17]

The gold standard method for diagnosing IHB is to measure TSB levels. Bilirubin values should be evaluated using the bilirubin nomogram based on the patient's age in hours. Nomograms can be used to track bilirubin values over time. It helps us predict which patients may develop hyperbilirubinemia in the future hours and days. TSB is regarded as the gold standard, but obtaining a good sample is not always possible, and the procedure can be painful. An alternative approach is the CT method, which requires a small blood sample. This method is less painful and more convenient.

Another alternative is to use TcB to assess bilirubin levels on the skin's surface. There is also the advantage of not having to provide a blood sample. This method reduces the frequency of severe hyperbilirubinemia and rehospitalization for phototherapy treatment. However, this method may give unreliable results in newborns undergoing phototherapy or newborns with dark skin.^[28]

Neonates are commonly discharged early because of medical, social, and economic constraints. Shortening the duration of hospitalization in the neonatal period is correlated with the risk of rehospitalization due to hyperbilirubinemia. Early IHB diagnosis and treatment reduce morbidity. TcB measurement is frequently used to measure bilirubin levels for screening purposes to diagnose IHB. We compared TcB methods, a noninvasive, economical, and rapid method for early diagnosis, with serum and CT bilirubin, an invasive method.

TcB device is performed by spectral subtraction due to light absorption of bilirubin in isolation in capillary beds and subcutaneous tissue. TcB measurements are a fast, easy, noninvasive, and inexpensive method for detecting IHB.^[21,23]

Gunaseelan et al. compared TcB sternum measurements with TSB measurements in a study on 400 newborns >35 weeks and found a significant correlation between them.^[18] Similarly, Ho et al. found that TcB significantly correlates with TSB measurements in term or near-term neonates. TcB and TSB measurements were evaluated in a 997-term and near-term newborn sample. TcB showed 100% sensitivity and 56% specificity in the low- and intermediate-risk phototherapy groups, assuming the 75th percentile cut level of the Bhutani nomogram as the threshold.

Using above the 75th percentile for high-risk patients resulted in 86.7% sensitivity and 97.0% specificity.^[19] In our study, the groups that received treatment comprised patients who scored 75% or higher on the Bhutani nomogram. Therefore, this explanation might be attributed to the statistically significant difference in TSB and TcB values in our study. Conversely, Maisels et al. and Rubaltelli et al. found a decrease in sensitivity when TcB had high cutoff measurement levels.^[20,21]

Bhutani et al.^[24] and Kolman et al.^[25] also reported a significant correlation between TcB and TSB. Compared to these studies, the results of our study might be different since we just included the high-risk group for IHB.

A meta-analysis by Kate et al. showed a good correlation between TcB measurements before and during phototherapy and TSB in term and preterm newborns. They reported that the use of TcB before and during phototherapy was a reliable method; however, they stated that there is insufficient data to determine the safety of TcB after phototherapy. Therefore, they stated that for TcB to be the most reliable value during phototherapy, it should be measured on closed skin (the part of the body that does not receive phototherapy).^[26]

A limitation of our study is that it did not include premature newborns. Jegathesan et al. reported that the TcB method is a noninvasive and reliable approach for screening hyperbilirubinemia before phototherapy in preterm infants born at 33-35 weeks of gestation. However, because the TcB method underestimates bilirubin levels after phototherapy in newborns born at <33 gestational weeks, TSB measurements are recommended to be preferred in the clinical decision-making process. When dealing with preterm infants born before 33 weeks of gestational age, the TcB method should be used with caution because their TSB levels may be near the phototherapy threshold. After phototherapy has begun, clinical decisions should be made based on TSB measurements because most preterm newborns require phototherapy within the first or second day of life.^[27]

Increasing the number of patients, including newborns under 35 weeks of gestation, may impact the outcomes. Furthermore, our study only included the high-risk group based on the Bhutani nomogram so that the results may differ in the low- and intermediate-risk groups.

Study Limitations

In our study, pre- and post-mature neonates, black race, blood exchange, and normal vaginal birth patients were not included. The results may differ when included in the study.

CONCLUSION

Our study found a statistically significant difference between bilirubin values of TSB with TcB and CT methods both before and after treatment. Although TcB and CT methods can be used in IHB screening, we cannot recommend using them instead of the TSB measurement method in terms of treatment and follow-up.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Tokat Gaziosmanpasa University Ethics Committee (Date: 18.01.2024, Decision No: 83116987-038).

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

Referee Evaluation Process: Externally peer-reviewed.

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