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Original Article

The Impact of Ursodeoxycholic Acid on Indirect Hyperbilirubinemia in Infants Treated with Phototherapy: A Single-Blind Randomized Clinical Trial

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ABSTRACT

Background: Infantile jaundice affects up to 84% of term infants and is a primary cause of re-hospitalization. Phototherapy, the primary therapeutic intervention, may be associated with several complications. Therefore, employing additional compensatory therapies could reduce the duration of phototherapy and its adverse effects.

Methods: A single-blind randomized clinical trial was conducted on newborns with indirect hyperbilirubinemia (IHB) undergoing phototherapy. Patients were randomly assigned to two groups: the intervention group received phototherapy and 10mg/kg of Ursodeoxycholic acid, taken twice a day with water as the solvent, while the control group received phototherapy and a placebo (water). Total bilirubin levels were measured every 12 hours, and the groups were compared regarding the reduction duration of bilirubin <10 mg/dl and the duration of phototherapy. Data analysis used SPSS-V22 software, with statistical tests done at a significance level of <0.05.

Results: In total, 128 neonates, comprising 56 (43.8%) males and 72 (56.2%) females, with a mean gestational age of 39.02±0.86 weeks and a mean age of 2.77±1.45 days, were subjected to analysis. After 96 hours, the mean bilirubin level was 9.23±2.81 mg/dl overall, 7.27±2.12 mg/dl in the case group, and 10.91±2.17 mg/dl in the control group (p<0.001). The mean time to reach a bilirubin level below 10 mg/dl was 51.66±17.27 hours overall, 58.38±10.05 hours for the case group, and 62.61±14.39 hours for the control group (p<0.001).

Conclusion: Ursodeoxycholic acid (UDCA) effectively reduces elevated bilirubin levels and the duration of phototherapy when employed as an adjunct treatment to phototherapy in cases of infantile indirect hyperbilirubinemia.

Keywords: Hyperbilirubinemia, Jaundice, Phototherapy, Ursodeoxycholic acid

Introduction

Infantile jaundice affects approximately 84% of term infants and ranks among the primary causes of re-hospitalization during infancy. Severe hyperbilirubinemia, defined as a total bilirubin level exceeding 20 mg/dl, may lead to kernicterus and enduring neurodevelopmental delays in less than 2% of term neonates (1).

More commonly, phototherapy proves beneficial in lowering bilirubin levels; nonetheless, it may be associated with certain complications and drawbacks, including high costs, occlusion of the infant's eyes, separation between mother and infant, retinal degenerative changes, electrolyte imbalances, bronze baby syndrome, and potentially life-threatening thermal fluctuations (2, 3). Consequently,

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employing supplementary and complementary therapies may mitigate the duration of phototherapy and its ensuing adverse effects (4, 5). Previous studies have demonstrated the advantageous effects of activated charcoal, probiotics, Di-penicillamine, phenobarbital, metalloporphyrin, and clofibrate in the treatment of indirect hyperbilirubinemia by reducing the duration of phototherapy; nevertheless, specific interventions have been accompanied by increased drowsiness, reduced breastfeeding, dehydration, and neurological deficits (6, 7). Consequently, there is a need to explore medications with fewer adverse effects.

Ursodeoxycholic acid (UDCA) is a biliary acid extensively employed in the treatment of cholestatic liver diseases through three principal mechanisms: protecting cholangiocytes against the cytotoxicity of hydrophobic biliary acids, stimulating hepatobiliary secretions, and safeguarding hepatocytes from biliary acid-related apoptosis (8, 9). The anti-cholestatic effects of UDCA have been established in conditions such as cystic fibrosis liver disease, familial intrahepatic cholestasis, and chronic graft-versus-host disease (10). UDCA is well-tolerated in pediatric patients and associated with limited complications, including gastrointestinal side effects, bloating, diarrhea, and allergic reactions (9, 11, 12).

A human study has confirmed the effectiveness of UDCA in reducing indirect bilirubin levels in infants undergoing phototherapy, as well as shortening the duration of phototherapy and hospitalization (13). Another trial by Mirzarahimi et al. showed that adding UDCA provided no significant difference compared to phototherapy alone (14).

Although some previous studies mentioned the positive effect of UDCA on neonatal hyperbilirubinemia (15-17), there is no consensus on its routine use. Therefore, it is not prescribed as an adjunct treatment in neonatal wards, and further studies are needed.

Due to the paucity of sufficient evidence, our objective was to assess the effects of UDCA on reducing indirect bilirubin levels, the duration of phototherapy, and the hospitalization period within a single-blind randomized clinical trial.

Methods

Study Design

This single-blind randomized clinical trial was conducted between March 2022 and March 2023 on neonates presenting with indirect hyperbilirubinemia and undergoing phototherapy treatment at Sayyad Shirazi Hospital in Gorgan, Iran. Participants were randomly allocated into two equally sized groups: the intervention group (n = 59) and the control group (n = 69). The study was carried out in strict adherence to the Consolidated Standards of Reporting Trials (CONSORT) Statement (18).

Participants and Setting

The protocol for this study was duly registered with the ethics committee of Golestan University of Medical Sciences in Gorgan, Iran. Parents were provided with a comprehensive explanation of the study's procedures and the potential adverse effects associated with it, following which they provided their informed consent by signing a consent form. Figure 1 presents a flowchart illustrating the study's progression.

Our study encompassed neonates whose characteristics met specific criteria, including birthweights ranging from 2500 to 4000 grams, exclusive breastfeeding, gestational ages within the 38 to 41-week range, age exceeding 24 hours, total bilirubin levels between 14 and 20 mg/dl, and direct bilirubin levels less than 2 mg/dl. Patients with ABO or Rh incompatibility, Glucose 6-phosphate (G6PD) deficiency, direct hyperbilirubinemia, septicemia, Crigler-Najjar disease, Gilbert syndrome, hypo- or hyperthyroidism, liver diseases, and neonate born to diabetic mothers were excluded from the study. Furthermore, the intervention was terminated if patients experienced adverse effects such as diarrhea, fever, cough, or other flu-like symptoms.

Sample Size

According to the study of Honar et al. (13), based on the primary outcome of the study, which was the total bilirubin level of the control group (9.8±1.1 mg/dl), using G power version 3.0.10 software with a confidence level of 95% and a test power of 80% in each group, at least 126 patients were required, which was expanded to 130 patients (65 people in each group) considering a 10% dropout.

$$n = \frac{\{Z_{1-\frac{\alpha}{2}}\sqrt{2\bar{P}(1-\bar{P})} + Z_{1-\beta}\sqrt{P_1(1-P_1)+P_2(1-P_2)}\}^2}{(P_1-P_2)^2}$$

Intervention and Outcomes

Neonate in the intervention group received phototherapy and a dose of 10mg/kg of Ursodeoxycholic acid (UDCA, Abidi Co., Tehran,

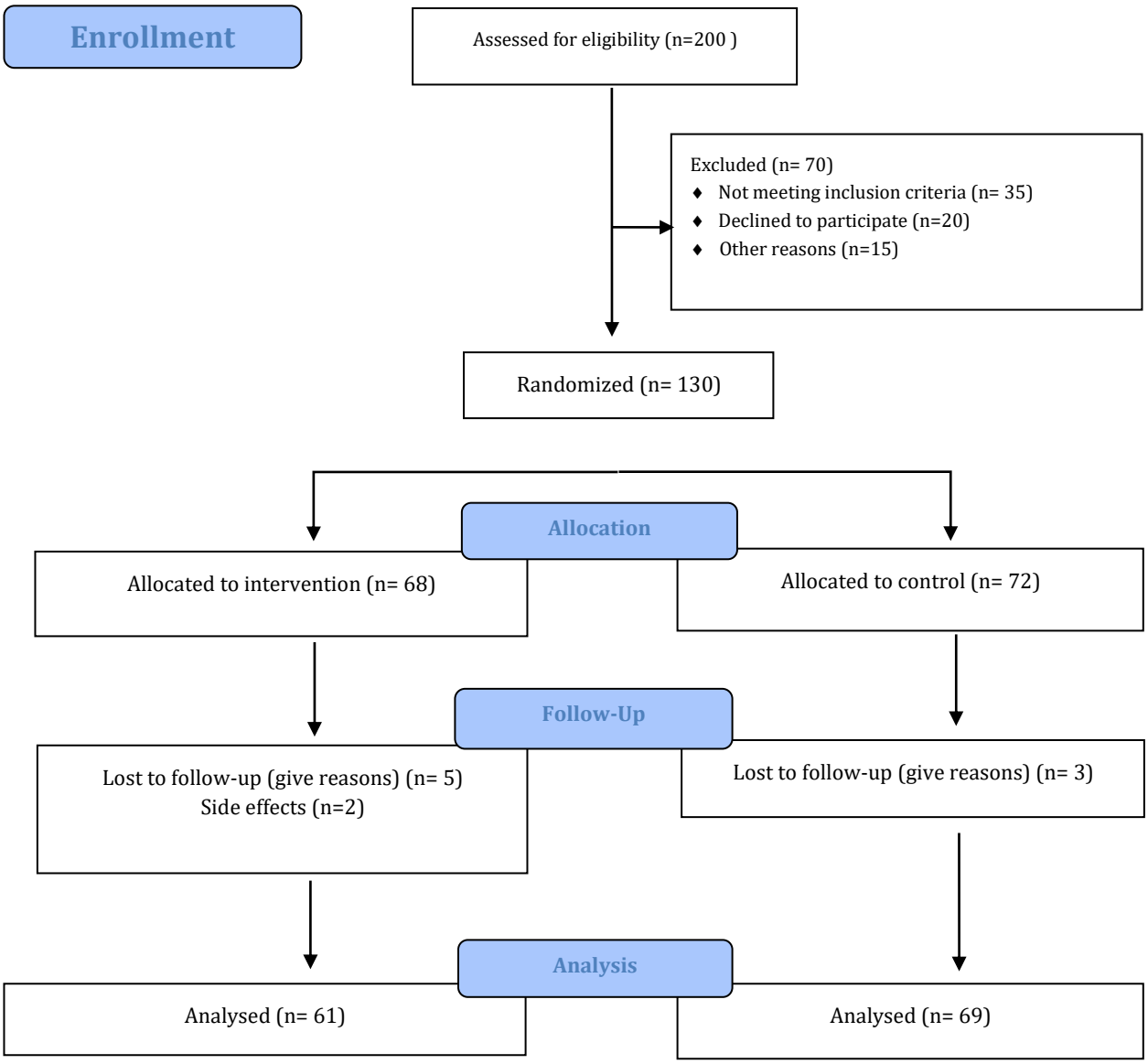

Figure 1. Study flowchart

Iran) in two doses per day (q 12 hours), utilizing water as the solvent. UDCA is obtainable in capsule form, with dosages of 300 mg and 250 mg, which are water-soluble. Patients in the control group received phototherapy and a placebo of water. Phototherapy was administered through fluorescent daylight lamps positioned 30 to 35 cm above the neonates. The eyes and genital areas of the neonates were shielded during the phototherapy sessions. On the first day of hospitalization, comprehensive data including medical history, physical examination findings, levels of total and direct bilirubin, reticulocyte count, Coomb's test results, G6PD levels, complete blood count (CBC), and Rh status were meticulously recorded in a pre-designed checklist. Furthermore, total bilirubin levels were assessed every 12 hours using the Diazo method (The most widely used method for unconjugated bilirubin concentration determination is the Diazo method, which measures the color of azobilirubin), and phototherapy was discontinued when the total bilirubin level fell below 10 mg/dl. The comparison of total bilirubin levels at different time points, the time required to reach a total bilirubin level below 10 mg/dl, and the duration of phototherapy was conducted between the two study groups.

- Primary outcome variable: Reduction in serum bilirubin.
- Secondary outcome variables: Reduction in the duration of hospital stay and reduction in the duration of phototherapy.

Randomization and Blinding

Neonates were allocated into two equal groups using a permuted balanced block randomization method with a block size of 4, ensuring allocation concealment. To determine the group allocation, one of the following blocks was randomly selected by rolling a die, and the remaining samples were divided into two groups following the sample order. Group A received Ursodeoxycholic, while Group B received a placebo.

The allocation sequences were as follows:

1. ABAB
2. BABA
3. BBAA
4. ABBA
5. AABB
6. ABAB

In this study, the parents and the outcomes assessor remained blinded to the patients' group assignments.

Statistical Analysis

The data were analyzed using SPSS software (version 16, SPSS Inc., Chicago, IL). The Chi-square test was employed to compare categorical variables between the two groups. Numerical variables were assessed using the Student's t-test or One-way Analysis of Variance (ANOVA). Paired categorical data were evaluated using the McNemar-Bowker test, while paired numerical data were assessed using the paired-sample t-test. A p-value of less than 0.05 was deemed statistically significant.

Ethical approval

All parents or legal guardians of neonates provided written informed consent after receiving a comprehensive explanation of the study. They retained the autonomy to decline participation or to withdraw from the study at any point. This research received ethical approval from the Ethics Committee of Golestan University of Medical Sciences (Ethics Committee reference code: IR.Goums.REC.1398.321), and it was additionally registered in the Iranian Registry of Clinical Trials under the identifier IRCTID: 20210222050449N1.

Results

Demographic Findings

Finally, 128 neonates underwent analysis, comprising 56 (43.8%) males and 72 (56.2%) females. The mean gestational age was 39.02 ± 0.86 weeks, ranging from a minimum of 38 to a maximum of 41 weeks, and the mean age at the time of analysis was 2.77 ± 1.45 days, with a minimum of one day and a maximum of six days. Additionally, the patients exhibited a mean weight of 3255.16 ± 525.34 grams, with weights ranging from a minimum of 2500 grams to a maximum of 4000 grams. Of the neonates, 56 (43.8%) were male, and 72 (56.2%) were female. Please refer to Table 1 for a summary of the demographic data.

Side Effect

During this study, five neonates from the case group were lost to follow-up, with two experiencing side effects. Specifically, two neonates developed diarrhea. Consequently, these patients were excluded from the intervention.

Bilirubin Levels of Newborns at 0-12-24-48 Hours After Treatment

Upon admission, the mean bilirubin level in the case group was 17.86 ± 1.5 mg/dl; in the control group, it was 17.87 ± 1.53 mg/dl ($p = 0.986$, Mann-Whitney test). After twelve hours of intervention, the mean bilirubin level for all patients was 16.70 ± 1.29 mg/dl, with 16.46 ± 1.38 mg/dl in the case group and 16.91 ± 1.18 mg/dl in the control group ($p = 0.044$, Mann-Whitney test). At the 24-hour evaluation, the mean bilirubin level was 14.09 ± 2.06 mg/dl for all patients, 13.17 ± 2.23 mg/dl in the case group, and 14.78 ± 1.35 mg/dl in the control group ($p < 0.001$, Mann-Whitney test). Forty-eight hours after the intervention, the mean bilirubin level stood at 9.23 ± 2.81 mg/dl for all patients, 7.27 ± 2.12 mg/dl in the case group, and 10.91 ± 2.17 mg/dl in the control group ($p < 0.001$, Mann-Whitney test).

Table 1. Demographic characteristics of study individuals

Variable	Total (N=128)	Case (N=59)	Control (N=69)	p-value*
Gestational age, weeks (mean ±SD)	39.02±0.86	39.03±0.87	39±0.86	0.825
Age, days (mean ±SD)	2.77±1.45	2.71±1.41	2.81±1.49	0.70
Age, hours (mean ±SD)	74.47±35.71	73.95±35.35	74.91±36.27	0.88
Weight, grams (mean ±SD)	3255.16±525.34	3279.15±528.40	3234.64±525.70	0.63
Gender, male, N(%)	56(43.8%)	25(42.4%)	31(44.9%)	0.73

* Independent Samples T Test

Duration of Serum Bilirubin Reduction to Less Than 10 mg/dl

Based on the Mann-Whitney test, The mean time required to attain a serum bilirubin level of less than 10 mg/dl was 51.66 ± 17.27 hours for all patients, 58.38 ± 10.05 hours for the case group, and 62.61 ± 14.39 hours for the control group ($p < 0.001$, Figure 2).

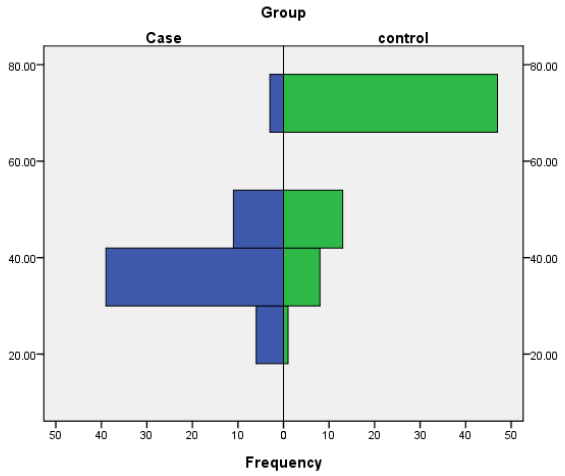

Figure 2. Mean time (hours) needed to achieve a bilirubin level of less than 10 mg/dl

Discussion

In this study, the intervention group (UDCA and phototherapy) experienced a significantly more significant reduction in bilirubin levels than the control group (placebo and phototherapy). This finding aligns with previous studies and can be attributed to increased biliary flow, reduced reabsorption of biliary acids, and decreased bilirubin concentration (19). None of the infants in the intervention group experienced an increase or consolidation of bilirubin levels, while a few infants in the control group exhibited some degree of bilirubin level increase or consolidation despite phototherapy. Additionally, the intervention group showed a significantly more significant reduction in bilirubin levels at all assessment points (12, 24, and 48 hours after the intervention), consistent with previous studies (15, 17, 20, 21). All these studies reported that UDCA was effective in accelerating the reduction of total bilirubin levels in neonates with unconjugated hyperbilirubinemia under phototherapy but had no effect on direct bilirubin levels. In some of these studies, the percentage of total bilirubin reduction in the intervention group was 1.5 times that of the control group (15, 20). In a similar study, it was found that the decrease in total bilirubin was greater in the first 8 hours of receiving UDCA (17).

Two systematic reviews demonstrated the efficacy of UDCA in neonatal IHB (22, 23). Nevertheless, in another trial by Mirzarahimi et al., the addition of UDCA made no significant difference compared to phototherapy alone (14). Akefi et al. showed that despite the significant decrease in the bilirubin level at 12 and 24 hours after treatment in the UDCA-receiving group, phototherapy duration was not different in the two groups(16).

Furthermore, the mean duration of phototherapy was significantly shorter in the intervention group compared to the control group, supported by previous findings and attributed to the adjunct effect of UDCA (13, 17, 24). A decrease in bilirubin levels negatively affects the efficacy of phototherapy due to the transmission of secreted unstable bilirubin isomers (photo bilirubin) to the intestines and their reabsorption through the enterohepatic cycle (21, 25). It is worth noting that UDCA inhibits the enterohepatic cycle (23). Phototherapy is generally considered a safe therapeutic method and should continue until jaundice is entirely resolved. However, the economic burden of hospitalizing mothers and infants has not been evaluated (21). In addition to evident expenses, other less obvious disadvantages, such as mental fatigue or separation from older siblings' care, may be mitigated by reducing the duration of hospitalization.

Moreover, a few complications were recorded in infants receiving UDCA (2.9%). Similar studies have also reported the well-tolerated nature of this medication, with only limited side effects reported in some cases (16, 26).

The present study had some limitations, the most significant being its coinciding with the COVID-19 pandemic, resulting in limited participant acceptance and, consequently, difficulties in achieving a sufficient sample size.

Conclusion

In conclusion, phototherapy remains a suitable and recommended therapeutic approach for non-complicated infantile hyperbilirubinemia, as endorsed by healthcare professionals. Adding UDCA as an adjunct therapy to phototherapy can expedite the reduction of bilirubin levels and reduce the required duration of phototherapy. This intervention could enhance parental compliance with phototherapy by decreasing hospitalization

duration and treatment costs. Further multicenter studies with larger sample sizes are recommended to evaluate the effectiveness of UDCA in combination with phototherapy for treating infantile hyperbilirubinemia.

Acknowledgments

None.

Conflicts of interest

The authors declare there is no conflict of interest.

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