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**Conjugated hyperbilirubinemia presenting in first fourteen days in term neonates**

Chiou FK *et al.* Neonatal conjugated hyperbilirubinemia within 14 d

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**Abstract**

***AIM***

To describe the etiology and characteristics of early-onset conjugated hyperbilirubinemia (ECHB) presenting within 14 d of life in term neonates.

***METHODS***

Retrospective review was performed of term infants up to 28-d-old who presented with conjugated hyperbilirubinemia (CHB) at a tertiary center over a 5-year period from January 2010 to December 2014. CHB is defined as conjugated bilirubin (CB) fraction greater than 15% of total bilirubin and CB greater or equal to 25 µmol/L. ECHB is defined as CHB detected within 14 d of life. ‘Late-onset’ CHB (LCHB) is detected at 15-28 d of life and served as the comparison group.

***RESULTS***

Total of 117 patients were recruited: 65 had ECHB, 52 had LCHB. Neonates with ECHB were more likely to be clinically unwell (80.0% *vs* 42.3%, *P* < 0.001) and associated with non-hepatic causes (73.8% *vs* 44.2%, *P* = 0.001) compared to LCHB. Multifactorial liver injury (75.0%) and sepsis (17.3%) were the most common causes of ECHB in clinically unwell infants, majority (87.5%) had resolution of CHB with no progression to chronic liver disease. Inborn errors of metabolism were rare (5.8%) but associated with high mortality (100%) in our series. In the subgroup of clinically well infants (*n* = 13) with ECHB, biliary atresia (BA) was the most common diagnosis (61.5%), all presented initially with normal stools and decline in total bilirubin but with persistent CHB.

***CONCLUSION***

Secondary hepatic injury is the most common reason for ECHB. BA presents with ECHB in well infants without classical symptoms of pale stools and deep jaundice.

**Key words:** Conjugated hyperbilirubinemia; Biliary atresia; Neonatal jaundice; Cholestasis; Direct hyperbilirubinemia

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**Core Tip:** Conjugated hyperbilirubinemia (CHB) is not routinely checked before 14-21 d of life, hence incidence and etiology of early-onset CHB (ECHB) before 14 d are not well-documented. Nearly three-quarters of ECHB have non-hepatic cause and are expected to recover with supportive treatment, while biliary atresia and metabolic disorders are important etiologies associated with significant morbidity. In our study, BA presenting before 14 d were detected solely from low levels of CHB without pale stools or worsening jaundice. Further studies are needed to determine if CHB screening before 14 d would lead to improved detection and outcome in neonatal liver disorders.

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**INTRODUCTION**

Conjugated hyperbilirubinemia (CHB) in a neonate signifies an underlying hepatobiliary dysfunction. A significant proportion of neonates with CHB do not have a primary liver disease[1,2]. According to current recommendations, serum conjugated bilirubin (CB) is checked when neonatal jaundice is prolonged beyond 14–21 d, prior to that only total bilirubin (TB) is checked[3,4]. The detection of CHB presenting before 14 d of life is usually triggered by specific clinical situations, therefore the real incidence and etiology of CHB in neonates below 14 d are unknown.

Even with well-established guidelines for the screening of neonatal CHB, actual referral for evaluation of CHB is frequently delayed to beyond 45 to 60 d of age[5-7]. Substantial observational evidence show that earlier diagnosis and surgical repair of biliary atresia (BA) result in better outcomes[8-11]. Early diagnosis of many of the other cholestatic conditions may also lead to improved outcomes[4]. Studies on infants with liver diseases including BA have shown that CB is often elevated in the first week of life[12-14]. Researchers have also found that CB level performed during the early newborn period is a useful “screening tool” for liver disorders especially biliary atresia[15].

We studied term newborns with CHB within 14 d of life, aiming to describe the etiology, clinical features and outcome in this poorly studied group, and to find out how they compare to those presenting with CHB between 15 to 28 d of life. To date, our study is the first to address CHB in full-term infants aged below 14 d.

**MATERIALS AND METHODS**

Retrospective data was collected from consecutive term infants with CHB below 28 d of age within a 5-year period from January 2010 to December 2014. Study was conducted at KK Women’s and Children’s Hospital which is the largest tertiary pediatric and neonatal facility in Singapore. The study was approved by Singhealth Centralised Institutional Review Board.

CHB is defined as CB fraction greater than 15% of TB, and CB ≥ 25 µmol/L[16-18]. We define “early-onset” as detection of CHB within 14 d of life (ECHB). Cases were identified through a search in the laboratory database using the inclusion criteria “conjugated bilirubin ≥ 25 µmol/L,” “conjugated bilirubin/total bilirubin > 15%,” “test performed at patient age ≤ 14 d.” Infants born at less than 36 wk gestation were excluded.

Consecutive term neonates presenting with CHB aged 15–28 d within the same period served as the comparison group. For the purpose of this study, this group presenting after 14 d of life is referred to as ‘late-onset’ CHB (LCHB).

CB was measured using an automated diazo dye reaction method from venous blood obtained by venipuncture in all patients. Blood samples were delivered immediately to the laboratory in covered specimen tubes to minimize the effect of light on the samples. Blood samples underwent an automated estimation of the hemolysis index, and samples that were found to be hemolysed based on established laboratory criteria were rejected, and repeat samples were taken.

Infants with CHB underwent a variety of investigations that included liver enzyme measurements, hepatobiliary ultrasonography, hepatobiliary imunodiacetic acid (HIDA) scan, liver biopsy, tests for inborn errors of metabolism (IEM), thyroid functions, bacterial cultures and viral serologies depending on the judgement of the treating physician. Surgical conditions such as BA and choledochal cysts were diagnosed from biochemical tests, radiologic findings and intra-operative cholangiography. IEM were diagnosed if confirmed report of an abnormality was found on appropriate testing. Multifactorial liver injury (MLI, hypoxic-ischemic or toxic) was defined in our study as secondary hepatic insult in an unwell neonate with any combination of the following: severe cadiorespiratory instability, hepatoxic medications and parenteral nutrition. Sepsis was defined as infection in which a viral or bacterial agent was isolated, and the infection was the primary cause of illness in the child. CHB was categorized as idiopathic if no cause was identified.

Data on patient demography, clinical history, co-morbid conditions, drug history, clinical status at time of detection of CHB, laboratory parameters, radiologic investigations and histologic studies, final diagnoses as well as outcome were retrospectively obtained from medical records. An infant was classified as clinically unwell when the admitting physician documented that the infant appeared unwell.

Data analysis was performed using IBM SPSS Statistics for Windows, version 19 (IBM Corp, Armonk, NY, United States). Continuous variables were expressed as mean ± SD or median (25%-75% interquartile range). Categorical variables were expressed as number (proportion). Comparisons were performed using two sample *t*-test in normally distributed data with equal variance or Mann-Whitney U test when the assumptions of two sample *t*-test were not met. *χ*2 test or Fisher’s exact test was used to compare categorical variables. Statistical significance was set at *P* < 0.05.

**RESULTS**

Total of 117 neonates with CHB were included in the study. Sixty-five had ECHB, and 52 LCHB. Baseline characteristics and liver function tests at presentation are summarized in Table 1. There was a significant male preponderance in both groups, and higher proportion of clinically unwell neonates in ECHB.

Etiology of CHB was identified in about 93% and 60 % of cases in ECHB and LCHB groups respectively, rest were classified as idiopathic. Non-hepatic cause for CHB was 73.8% *vs* 44.2% (*P* = 0.001) in ECHB and LCHB respectively. MLI was an attributable cause of ECHB in 60%, followed by primary sepsis (13.8%) and BA (12.3%) (Table 2). In contrast, the most common cause found in LCHB was idiopathic (40.4%), followed by MLI (34.6%) and BA (9.6%). Factors associated with MLI in both ECHB and LCHB groups are summarized in Table 3.

There was a significantly higher proportion of unwell infants in ECHB group, 80.0% *vs* 42.3% in LCHB group (*P* < 0.001) (Tables 1 and 4). In the subgroup of patients who were clinically well within the ECHB group, BA was the most common diagnosis (61.5%), the remaining were idiopathic. The most common etiology/association found in well infants in the LCHB group was idiopathic (70.0%), followed by surgical causes (23.4%). No patient with BA was clinically unwell.

Out of the 65 patients with ECHB, 47 (72.3%) resolved within a mean period of 1.9 ± 1.4 mo with eventual normalization of liver tests, 8 (12.3%) had surgery for BA and 8 (12.3%) died. Five deaths were due to multi-organ failure and three due to IEM. In the subgroup of patients with ECHB due to non-hepatic causes (*n* = 48), 42 (87.5%) achieved complete resolution of CHB without progression to chronic liver disease. In comparison, in the LCHB group overall (*n* = 52), 41 (78.8%) had complete resolution, 7 (13.5%) underwent surgery for BA and choledochal cyst, 2 (3.8%) patients died, one due to IEM and the other died with multi-organ failure. Two patients from each group, ECHB and LCHB, were lost to follow-up. Death occurred in all 4 patients with IEM, three of them in the ECHB group (two mitochondrial disorders and one organic aciduria) and one in LCHB group with urea cycle defect. In both ECHB and LCHB groups, all patients with MLI who survived and all those with idiopathic CHB had complete resolution of liver dysfunction on follow-up.

The reasons for measuring serum CB in the well-looking ECHB cases were that atypical “bronze” appearance of skin (38.5%), screening at physician’s discretion (30.8%), antenatally detected hepatobiliary anomalies (15.4%) and non-specific symptoms such as vomiting, abdominal distension, respiratory distress and hypoglycemia (15.4%). In eight infants with biliary atresia who presented with ECHB, four had atypical “bronze” appearance, two had antenatally detected hepatobiliary anomalies, and two were screened on physicians’ discretion. None of these BA infants had acholic stools at presentation. They also had an initial declining trend of TB, reaching below 50% of initial values in 5 of them, while their CB remained persistently elevated.

**DISCUSSION**

CHB is often detected when infants are investigated for prolonged neonatal jaundice beyond 14–21 d of life[4]. Although less routinely encountered, neonatal CHB presenting within 14 d of life can pose considerable diagnostic and management challenges. In one study, the most common etiology of CHB (mean age 10 d) admitted to neonatal intensive care unit (NICU) was culture-proven sepsis (35.5%) and 30 out of 42 (71%) had non-hepatic cause[1]. In our study, the proportion of neonates with non-hepatic cause for CHB was similar (61%). However, the incidence of sepsis was much lower (10.3%), this difference is because 36% of neonates in that study were preterm requiring NICU care who were more likely to be predisposed to sepsis. Reported etiology of CHB differed depending upon age distribution, geographical region, type of study center and diagnostic approach[19]. We excluded preterm infants and focused on CHB in term neonates, including those who did not require hospitalization. Most studies on infantile cholestasis focus on BA but we did not find any study looking specifically into the clinical course of neonates with CHB aged below 14 d.

Similar to several other studies, MLI was an important etiology in our series and accounted for almost fifty percent[2,19-22]. Neonates are predisposed to MLI and cholestasis due to the relative immaturity of the hepatobiliary system, exacerbated by a wide variety of neonatal events such as hypoxia, prolonged fasting, parenteral nutrition, drug toxicity and sepsis[2-3,22-25]. Liver injury in such cases is part of multi-organ involvement. The severity and persistence of liver dysfunction depend on underlying disorders, and the dysfunction is usually reversible after resolution of the primary problem[21-23]. Standard intensive care management of the sick infant and close monitoring of liver function are the mainstays of treatment in these cases. In our study, CHB resolved without any long term liver complications in all the surviving infants with MLI and sepsis, majority of them (91%) recovered within 3 mo.

A significantly higher proportion of newborns who presented before 14 d were clinically unwell compared to those presenting later (80% *vs* 42%), (Table 4). As per guidelines, healthy infants below 14 d with jaundice are rarely tested for CB, potentially missing CHB in healthy patients and over-estimating the proportion of unwell patients. We observed that about three-quarters of clinically unwell CHB patients presenting within 14 d had non-hepatic cause for CHB. Importantly, no clinically unwell patient had BA (Table 4). The presence of IEM was an important risk factor for mortality. IEM have been reported to account for about 20% of all cases of neonatal cholestasis[16,19]. It is therefore recommended to maintain a high level of suspicion for IEM in unwell infants with CHB[26].

Excluding clinically unwell infants, the most common cause of ECHB is BA (61.5%). Notably all infants with BA had pigmented stools at this early stage. Prognosis of BA is dependent on timely diagnosis and surgical intervention. Despite data from BA case series suggesting presence of jaundice before 14 d[27-28], a significant proportion of cases are referred after 6 to 8 wk of life[5], and the age at which the Kasai operation is performed has not decreased over the years[8-11].

In our study, all patients with BA in the ECHB group had a significant initial decline of TB, and in 5 out of 8, TB fell by over 50% from presentation levels, reaching clinically undetectable levels (below 70 µmol/L). It can be argued that BA cases may initially have unconjugated hyperbilirubinemia, and CHB develops later. In our study the subset of infants with ECHB who were diagnosed to have BA continued to have persistently raised CB, and this observation was also seen in other studies[15,28]. Measuring CB in all patients with neonatal jaundice regardless of age, and investigating those with CHB could potentially discover BA at an earlier stage. A recent study examined the potential utility of newborn direct bilirubin measurements performed prior to 60 h of life when infants are still in the hospital as a screen for BA. Authors predicted sensitivity of 100%, based on 35 subjects with BA and predicted specificity of 98.2% based on 9102 subjects without BA[15].

A few indications to measure CB in well looking neonates below 14 d are antenatally detected hepatobiliary anomalies, pale stools, dark urine and bronze baby syndrome[29]. In our study approximately one-third developed bronze baby syndrome, 15% had antenatally detected hepatobiliary anomalies, while none had pale stools or dark urine. This highlights that even with good antenatal ultrasonogram and careful clinical evaluation, a significant proportion of ECHB can be missed.

Delayed detection of neonatal CHB and BA in particular is unlikely to be confined to lack of training and awareness of guidelines among healthcare providers, as despite having guidelines for over 2 decades, cases continue to be missed and treatment delayed[7]. This is likely to be due to subjectivity in assessment of jaundice. Firstly, it is difficult for parents and physicians to detect minimal jaundice. In addition, as shown in our study, the initial decline of TB may give a false reassurance and the well-looking infant may not be followed-up with blood tests[5]. Parents may also avoid clinic visits if the infant appears to be improving, this may be for economic reasons or to protect infants from the discomfort of venipuncture.

Hypothetically, if CB is checked with TB measurement during neonatal jaundice screening, or within 60 h of life in all infants[15], we believe liver disorders and BA can be detected earlier. However, there is no data on the cost-effectiveness of such an approach. It is worthwhile to study the increased economic and logistic burden that arises from over-investigating the self-resolving cases and weigh it against the benefits of earlier detection of CHB. We acknowledge that this approach is not applicable in centers relying on transcutaneous bilirubin (TcB) or in areas where BA prevalence is low. Mowat *et al*[6] discussed screening for CHB and suggested checking urine for conjugated bilirubin, its usefulness as an adjunctive test could be explored in scenarios where blood testing is deemed unnecessary and/or in units relying on TcB.

The main limitation of this study is the single-center retrospective data that could result in selection bias, particularly over-representation of unwell infants with ECHB and under-representation of untested well-looking infants with BA. Another limitation is the non-availability of liver biopsy data in all the cases which could potentially influence the accuracy of diagnosis. This study serves as a primer for prospective studies to evaluate the role of routine measurement of CB in neonatal jaundice and its impact on the outcomes of CHB.

In conclusion,non-hepatic etiology is the most common reason for ECHB in term neonates aged below 14 d. In clinically unwell neonates who do not have IEM, CHB is expected to resolve with supportive management.

BA is an important cause of ECHB in well-looking, jaundiced term infants; it is also an unlikely diagnosis in clinically unwell, jaundiced neonates. Low level of CHB is present in all cases of BA who had CHB tested prior to 14 d of life; large population-based studies may be able to provide the answer whether routine measurement of conjugated bilirubin in all neonates with jaundice regardless of age, may potentially lead to earlier detection of biliary atresia and other neonatal liver disorders.

**COMMENTS**

***Background***

Conjugated hyperbilirubinemia (CHB) in a neonate may be indicative of serious hepatobiliary pathology, such as biliary atresia (BA) or inborn errors of metabolism (IEM). Based on current guidelines, conjugated bilirubin (CB) is screened when neonatal jaundice persists beyond 14–21 d. Hence, incidence and etiology of neonatal CHB before 14 d are not well-defined. Published data suggest that diagnosis of neonatal liver diseases including BA is frequently delayed, and earlier detection can lead to improved outcomes for these infants.

***Research frontiers***

Early-onset CHB (ECHB) presenting in the first 14 d of life in neonates remain poorly-defined. At the time of writing, there is no other study looking specifically into the clinical course of term neonates presenting with ECHB. The results of this study may contribute to understanding the etiologies of ECHB in term infants and earlier detection/diagnosis of neonatal liver disorders.

***Innovations and breakthroughs***

This study shows that non-hepatic etiology is the most common reason for ECHB in term neonates aged below 14 d, particularly in the subgroup who are clinically unwell. IEM are rare but associated with high mortality. On the other hand, BA is an important cause of ECHB in well-looking, jaundiced term infants who may not exhibit classical symptoms and signs at this early stage, making the diagnosis of BA difficult if current guidelines are followed. Low level of CHB was found to be present in all cases of BA who had CHB tested prior to 14 d of life.

***Applications***

In clinically unwell infants with ECHB, if rare IEM are excluded early, majority of cases with non-hepatic causes are expected to resolve with supportive management without progression to chronic liver disease. However, BA should be suspected in well infants presenting with ECHB, even in the absence of pale stools or deep jaundice. This study serves as a primer for larger population-based studies to evaluate the cost-effectiveness of earlier screening for conjugated bilirubin before 14 d in term infants, and its impact on the outcome of neonatal liver disorders including BA.

***Terminology***

CHB is defined as conjugated bilirubin CB fraction greater than 15% of TB, and CB ≥ 25 µmol/L. ECHB is defined as CHB detected within 14 d of life.

***Peer-review***

This retrospective single-center study may contribute to early detection of the cause of conjugated hyperbilirubinemia in term infants.

**REFERENCES**

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| --- |
| 1 **Tiker F**, Tarcan A, Kilicdag H, Gurakan B. Early onset conjugated hyperbilirubinemia in newborn infants. *Indian J Pediatr* 2006; **73**: 409-412 [PMID: 16741326 DOI: 10.1007/BF02758562] |
| 2 **Jacquemin E**, Lykavieris P, Chaoui N, Hadchouel M, Bernard O. Transient neonatal cholestasis: origin and outcome. *J Pediatr* 1998; **133**: 563-567 [PMID: 9787700 DOI: 10.1016/S0022-3476(98)70070-8] |
| 3 **McKiernan PJ**. Neonatal cholestasis. *Semin Neonatol* 2002; **7**: 153-165 [PMID: 12208100 DOI: 10.1053/siny.2002.0103] |  |
| 4 **Moyer V**, Freese DK, Whitington PF, Olson AD, Brewer F, Colletti RB, Heyman MB; North American Society for Pediatric Gastroenterology, Hepatology and Nutrition. Guideline for the evaluation of cholestatic jaundice in infants: recommendations of the North American Society for Pediatric Gastroenterology, Hepatology and Nutrition. *J Pediatr Gastroenterol Nutr* 2004; **39**: 115-128 [PMID: 15269615 DOI: 10.1097/00005176-200408000-00001] |
| 5 **Mieli-Vergani G**, Howard ER, Portman B, Mowat AP. Late referral for biliary atresia--missed opportunities for effective surgery. *Lancet* 1989; **1**: 421-423 [PMID: 2563796 DOI: 10.1016/S0140-6736(89)90012-3] |  |
| 6 **Hussein M**, Howard ER, Mieli-Vergani G, Mowat AP. Jaundice at 14 days of age: exclude biliary atresia. *Arch Dis Child* 1991; **66**: 1177-1179 [PMID: 1952998 DOI: 10.1136/adc.66.10.1177] |
| 7 **Mowat AP**, Davidson LL, Dick MC. Earlier identification of biliary atresia and hepatobiliary disease: selective screening in the third week of life. *Arch Dis Child*1995; **72**: 90-92 [PMID: 7717750 DOI: 10.1136/adc.72.1.90] |  |
| 8 **Nio M**, Ohi R, Miyano T, Saeki M, Shiraki K, Tanaka K; Japanese Biliary Atresia Registry. Five- and 10-year survival rates after surgery for biliary atresia: a report from the Japanese Biliary Atresia Registry. *J Pediatr Surg* 2003; **38**: 997-1000 [PMID: 12861525 DOI: 10.1016/S0022-3468(03)00178-7] |
| 9 **Wadhwani SI**, Turmelle YP, Nagy R, Lowell J, Dillon P, Shepherd RW. Prolonged neonatal jaundice and the diagnosis of biliary atresia: a single-center analysis of trends in age at diagnosis and outcomes. *Pediatrics* 2008; **121**: e1438-e1440 [PMID: 18443020 DOI: 10.1542/peds.2007-2709] |  |
| 10 **Serinet MO**, Wildhaber BE, Broué P, Lachaux A, Sarles J, Jacquemin E, Gauthier F, Chardot C. Impact of age at Kasai operation on its results in late childhood and adolescence: a rational basis for biliary atresia screening. *Pediatrics* 2009; **123**: 1280-1286 [PMID: 19403492 DOI: 10.1542/peds.2008-1949] |
| 11 **Muraji T**. Early detection of biliary atresia: past, present &amp; future. *Expert Rev Gastroenterol Hepatol* 2012; **6**: 583-589 [PMID: 23061709 DOI: 10.1586/egh.12.37] |  |
| 12 **Keffler S**, Kelly DA, Powell JE, Green A. Population screening for neonatal liver disease: a feasibility study. *J Pediatr Gastroenterol Nutr* 1998; **27**: 306-311 [PMID: 9740202 DOI: 10.1097/00005176-199809000-00007] |
| 13 **Powell JE**, Keffler S, Kelly DA, Green A. Population screening for neonatal liver disease: potential for a community-based programme. *J Med Screen* 2003; **10**: 112-116 [PMID: 14561261 DOI: 10.1177/096914130301000303] |  |
| 14 **Davis AR**, Rosenthal P, Escobar GJ, Newman TB. Interpreting conjugated bilirubin levels in newborns. *J Pediatr* 2011; **158**: 562-565.e1 [PMID: 21074172 DOI: 10.1016/j.jpeds.2010.09.061] |
| 15 **Harpavat S**, Ramraj R, Finegold MJ, Brandt ML, Hertel PM, Fallon SC, Shepherd RW, Shneider BL. Newborn Direct or Conjugated Bilirubin Measurements As a Potential Screen for Biliary Atresia. *J Pediatr Gastroenterol Nutr* 2016; **62**: 799-803 [PMID: 26720765 DOI: 10.1097/MPG.0000000000001097] |  |
| 16 **Suchy FJ**. Neonatal cholestasis. *Pediatr Rev* 2004; **25**: 388-396 [PMID: 15520084 DOI: 10.1093/pch/9.10.700] |
| 17 **Gilmour SM**. Prolonged neonatal jaundice: When to worry and what to do. *Paediatr Child Health* 2004; **9**: 700-704 [PMID: 19688078 DOI: 10.1093/pch/9.10.700] |  |
| 18 **National Institute for Health and Care Excellence (NICE).** Formal assessment for the causes of neonatal hyperbilirubinaemia. In: NICE Clinical Guideline 98. Jaundice in newborn babies under 28 days. 2010. |
| 19 **Gottesman LE**, Del Vecchio MT, Aronoff SC. Etiologies of conjugated hyperbilirubinemia in infancy: a systematic review of 1692 subjects. *BMC Pediatr* 2015;**15**: 192 [PMID: 26589959 DOI: 10.1186/s12887-015-0506-5] |  |
| 20 **Vajro P**, Amelio A, Stagni A, Paludetto R, Genovese E, Giuffré M, DeCurtis M. Cholestasis in newborn infants with perinatal asphyxia. *Acta Paediatr* 1997; **86**: 895-898 [PMID: 9307175 DOI: 10.1111/j.1651-2227.1997.tb08619.x] |
| 21 **Stormon MO**, Dorney SF, Kamath KR, O'Loughlin EV, Gaskin KJ. The changing pattern of diagnosis of infantile cholestasis. *J Paediatr Child Health* 2001; **37**: 47-50 [PMID: 11168869 DOI: 10.1046/j.1440-1754.2001.00613.x] |  |
| 22 **İpek MŞ**, Aydın M, Zencıroğlu A, Gökçe S, Okumuş N, Gülaldı NC. Conjugated hyperbilirubinemia in the neonatal intensive care unit. *Turk J Gastroenterol*2013; **24**: 406-414 [PMID: 24557964 DOI: 10.4318/tjg.2013.0553] |
| 23 **Jacquemin E**, Saliba E, Blond MH, Chantepie A, Laugier J. Liver dysfunction and acute cardiocirculatory failure in children. *Eur J Pediatr* 1992; **151**: 731-734 [PMID: 1425791 DOI: 10.1007/BF01959078] |  |
| 24 **Khalil S**, Shah D, Faridi MM, Kumar A, Mishra K. Prevalence and outcome of hepatobiliary dysfunction in neonatal septicaemia. *J Pediatr Gastroenterol Nutr*2012; **54**: 218-222 [PMID: 21873892 DOI: 10.1097/MPG.0b013e318233d33d] |
| 25 **Rangel SJ**, Calkins CM, Cowles RA, Barnhart DC, Huang EY, Abdullah F, Arca MJ, Teitelbaum DH; 2011 American Pediatric Surgical Association Outcomes and Clinical Trials Committee. Parenteral nutrition-associated cholestasis: an American Pediatric Surgical Association Outcomes and Clinical Trials Committee systematic review. *J Pediatr Surg* 2012; **47**: 225-240 [PMID: 22244423 DOI: 10.1016/j.jpedsurg.2011.10.007] |  |
| 26 **Chakrapani A**, Cleary MA, Wraith JE. Detection of inborn errors of metabolism in the newborn. *Arch Dis Child Fetal Neonatal Ed* 2001; **84**: F205-F210 [PMID: 11320051 DOI: 10.1136/fn.84.3.F205] |
| 27 **Mowat AP**, Psacharopoulos HT, Williams R. Extrahepatic biliary atresia versus neonatal hepatitis. Review of 137 prospectively investigated infants. *Arch Dis Child* 1976; **51**: 763-770 [PMID: 1087549 DOI: 10.1136/adc.51.10.763] |  |
| 28 **Harpavat S**, Finegold MJ, Karpen SJ. Patients with biliary atresia have elevated direct/conjugated bilirubin levels shortly after birth. *Pediatrics* 2011; **128**: e1428-e1433 [PMID: 22106076 DOI: 10.1542/peds.2011-1869] |
| 29 **De Luca D**, Picone S, Fabiano A, Paolillo P. Images in neonatal medicine. Bronze baby syndrome: pictorial description of a rare condition. *Arch Dis Child Fetal Neonatal Ed* 2010; **95**: F325 [PMID: 20530102 DOI: 10.1136/adc.2010.185207] |  |

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**Table 1 Baseline clinical characteristics and biochemical indices at onset of conjugated hyperbilirubinemia**

|  |  |  |  |
| --- | --- | --- | --- |
| **Baseline characteristics** | **ECHB****(*n* = 65, %)** | **LCHB****(*n* = 52, %)** | ***P* value** |
| Ethnic originChineseMalayIndianOthers | 34 (52.3%)15 (23.1%)8 (12.3%)8 (12.3%) | 27 (51.9%)18 (34.6%)4 (7.7%)3 (5.8%) | 0.547 |
| Male gender | 38 (58.5%)  | 40 (76.9%) | 0.035 |
| Gestational age (wk) | 38 (37–39) | 38 (37–39) | 0.303 |
| Birth weight (g) | 2918 (2570–3245) | 3068 (2753–3416) | 0.114 |
| Apgar at 1 min at 5 min | 9 (6–9)9 (8–9) | 9 (9–10)9 (9–10) | 0.2170.134 |
| Cesarean section | 25 (38.5%) | 14 (26.9%) | 0.190 |
| Clinically ill status on presentation | 52 (80.0%) | 22 (42.3%) | < 0.001 |
| LFT (at diagnosis)Total bilirubin (µmol/L)Conjugated bilirubin (µmol/L)Conjugated fraction (%)ALP (IU/L)ALT (IU/L)AST (IU/L)GGT (IU/L) | 147 (100–201)46 (32–65)35.7 (24.0–51.4)160 (119–261)20 (13–42)35 (26–75)142 (74–334) | 120 (91–163)38 (30–74)37.4 (26.3–61.5)322 (238–418)23 (16–32)35 (25–52)199 (131–273) | 0.0330.3100.159< 0.0010.3770.5120.045 |

ECHB: Early-onset conjugated hyperbilirubinemia; LCHB: “Late-onset” conjugated hyperbilirubinemia; LFT: Liver function test; ALP: Alkaline phosphatase; ALT: Alanine transaminase; AST: Aspartate transaminase; GGT: Gamma-glutamyl transferase.

**Table 2 Comparison of causes between early-onset conjugated hyperbilirubinemia and ‘late-onset’ conjugated hyperbilirubinemia groups** ***n* (%)**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Etiology** | **ECHB (*n* = 65)**  | **LCHB (*n* = 52)**  | **total (*n* = 117)**  | ***P* value** |
| Non-surgical causes | 56 (86.2) | 45 (86.5) | 101 (86.3) | 0.962 |
| Multifactorial liver injury | 39 (60.0) | 18 (34.6) | 57 (48.7) | 0.007 |
| Sepsis | 9 (13.8) | 3 (5.8) | 12 (10.3) | 0.154 |
| Inborn errors of metabolism | 3 (4.6) | 1 (1.9) | 4 (3.4) | 0.428 |
| CMV infection | 0 (0) | 2 (3.8) | 2 (1.7) | 0.112 |
| Idiopathic | 5 (7.7) | 21 (40.4) | 26 (22.2) | < 0.001 |
| Surgical causes | 9 (13.8) | 7 (13.5) | 16 (13.7) | 0.952 |
| Biliary Atresia | 8 (12.3) | 5 (9.6) | 13 (11.1) | 0.647 |
| Choledochal cyst | 1 (1.5) | 2 (3.8) | 3 (2.6) | 0.435 |
| Non-hepatic causes† | 48 (73.8%) | 23 (44.2%) | 71 (61%) | 0.001 |

ECHB: Early-onset conjugated hyperbilirubinemia; LCHB: ‘Late-onset’ conjugated hyperbilirubinemia; CMV: Cytomegalovirus.

**Table 3 Factors associated with multifactorial liver injury** ***n* (%)**

|  |  |  |  |
| --- | --- | --- | --- |
| **Factors associated with multifactorial liver injury** | **ECHB****(*n* = 39)**  | **LCHB****(*n* = 18)**  | **Total** **(*n* = 57)**  |
| Antibiotics | 38 (97.4) | 18 (100.0) | 56 (98.2) |
| Parenteral nutrition | 35 (89.7) | 16 (88.9) | 51 (89.5) |
| Sedatives/Opioid | 29 (74.4) | 14 (77.8) | 43 (75.4) |
| Mechanical ventilation | 26 (66.7) | 12 (66.7) | 38 (66.7) |
| Inotropic support | 23 (59.0) | 9 (50.0) | 32 (56.1) |
| Recent surgery | 20 (51.3) | 12 (66.7) | 32 (56.1) |
| PPHN | 19 (48.7) | 4 (22.2) | 23 (40.4) |
| Intestinal obstruction | 13 (33.3) | 7 (38.9) | 20 (35.1) |
| Congenital heart disease | 12 (30.8) | 4 (22.2) | 16 (28.1) |
| HFOV | 11 (28.2) | 3 (16.7) | 14 (24.6) |
| Pneumothorax | 8 (20.5) | 1 (5.6) | 9 (15.8) |
| CDH | 5 (12.8) | 4 (22.2) | 9 (15.8) |
| MAS | 6 (15.4) | 2 (11.1) | 8 (14.0) |
| Renal impairment | 6 (15.4) | 2 (11.1) | 8 (14.0) |
| Seizures/ Anti-epileptic | 4 (10.3) | 2 (11.8) | 6 (10.7) |
| Perinatal asphyxia | 3 (7.7) | 3 (16.7) | 6 (10.5) |
| Intracranial haemorrhage | 2 (5.1) | 1 (5.6) | 3 (5.3) |
| Trisomy 21 | 2 (5.1) | 1 (5.6) | 3 (5.3) |
| ECMO | 2 (5.1) | 1 (5.6) | 3 (5.3) |
| Turner’s syndrome | 1 (2.6) | 0 | 1 (1.8) |
| Trisomy 18 | 1 (2.6) | 0  | 1 (1.8) |

ECHB: Early-onset conjugated hyperbilirubinemia; LCHB: ‘Late-onset’ conjugated hyperbilirubinemia; PPHN: Persistent pulmonary hypertension of the newborn; HFOV: High frequency oscillatory ventilation; CDH: Congenital diaphragmatic hernia; MAS: Meconium aspiration syndrome; ECMO: Extra-corporeal membrane oxygenation.

**Table 4 Comparison of causes of early-onset conjugated hyperbilirubinemia and “late-onset” conjugated hyperbilirubinemia between subgroups of clinically well and unwell infants** ***n* (%)**

|  |  |  |
| --- | --- | --- |
|  | **ECHB (*****n* = 65)** | **LCHB (*n* = 52)** |
| **Unwell (*n* = 52)**  | **Well (*n* = 13)**  | **Unwell (*n* = 22)**  | **Well (*n* = 30)**  |
| Non-surgical causes |  |  |  |  |
| Multifactorial liver injury | 39 (75.0) | 0 (0) | 18 (81.8) | 0 (0) |
| Sepsis | 9 (17.3) | 0 (0) | 3 (13.6) | 0 (0) |
| Inborn errors of metabolism | 3 (5.8) | 0 (0) | 1 (4.5) | 0 (0) |
| CMV infection | 0 (0) | 0 (0) | 0 (0) | 2 (6.7) |
| Idiopathic | 0 (0) | 5 (38.5) | 0 (0) | 21 (70.0) |
| Surgical causes |  |  |  |  |
| Biliary atresia | 0 (0) | 8 (61.5) | 0 (0) | 5 (16.7) |
| Choledochal cyst | 1 (1.9) | 0 (0) | 0 (0) | 2 (6.7) |

ECHB: Early-onset conjugated hyperbilirubinemia; LCHB: “Late-onset” conjugated hyperbilirubinemia; CMV: Cytomegalovirus.