Differential Effects of Later Cord Clamping on Bilirubin Levels in Normal and Diabetic Pregnancies

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Research Article

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Abstract

Purpose: To investigate the effect of later cord clamping on bilirubin levels and phototherapy rates in normal pregnancy term neonates and diabetic pregnancy neonates.

Methods: This is a prospective study that enrolled pregnant women without pregnancy complications and those with diabetes according to the criteria. The subjects were randomized in a 1:1 ratio to either immediate cord clamping or later cord clamping. The final data analysis involved 132 cases of diabetic pregnancy and 129 cases of normal pregnancy. The main outcomes were neonatal the transcutaneous bilirubin values in 2-4 days postpartum and the phototherapy rate.

Results: In diabetic pregnancy, neonatal bilirubin levels on the 2-4 day postpartum and phototherapy rates were significantly higher in later cord clamping group than in immediate cord clamping group, while in normal pregnancy, there was no statistical difference in bilirubin values and phototherapy rates between different clamping methods. After receiving later cord clamping, bilirubin levels on the third postnatal day and the rate of neonates needing phototherapy were higher in the diabetic pregnancy group than in the normal pregnancy group.

Conclusions: Later cord clamping increases the risk of jaundice in newborns whose mothers have diabetes, but has no effect on newborns with normal pregnancy.

Trial registration: ClinicalTrials.gov: NCT04369313; date of registration: April 27, 2020 (retrospectively registered)

What Is Known

- Later cord clamping has significant benefits for newborns by increasing neonatal hemoglobin levels and reducing the risk of neonatal anemia, etc.
- Later cord clamping may lead to neonatal hyperemia, erythrocytosis, and hyperbilirubinemia, which increases the risk of neonatal jaundice.

what is new:

- This is the first to describe the differential effect of later umbilical cord clamping on neonatal jaundice between normal pregnancies and diabetic pregnancies.
- Newborns of diabetic mothers who received later cord clamping had a significantly increased risk of jaundice compared with normal pregnancy.

Introduction

Later cord clamping (LCC) has significant benefits for newborns, especially preterm infants, by increasing neonatal hemoglobin levels, reducing required fluid intake for newborns, and reducing the risk of neonatal anemia, necrotizing enterocolitis, and ventricular bleeding[1–3]. The American College of obstetricians and Gynecologists recommends that preterm infants delivered vaginally receive a delay of cord clamping for at least 30s[4]. However, some researchers reported that excessive placental blood transport through LCC caused hypervolemia, erythrocytosis, hyperlipemia, and hyperbilirubinemia in neonates, thereby resulting in an elevated

likelihood of neonatal jaundice[5–7]. But other studies found no effect of LCC on neonatal bilirubin levels and phototherapy rates and there was no significant increase in the rate requiring partial exchange transfusion, although it can increase the rate of erythrocytosis[8, 9].

Diabetes, caused by impaired glucose metabolism, is one of the most common metabolic disorders, accounting for 3-10% of all pregnancies[10]. There are three main types of diabetes, of which gestational diabetes accounts for 88% of diabetics, and 4% and 8% of those who had type 1 and type 2 diabetes before pregnancy, respectively[10]. It has been found that any type of gestational diabetes is associated with an increased risk of neonatal hypoglycemia, erythrocytosis, jaundice, phototherapy rates, and respiratory distress syndrome compared to normal pregnancy[10–12]. In addition, LCC has been found to significantly increase bilirubin levels and phototherapy rates in neonates of diabetic mothers [13]. However, the difference in the impact of LCC on neonatal jaundice between normal and diabetic pregnancies is unknown.

The purpose of this trial was to investigate the differential effects of LCC on day 2-4 transcutaneous bilirubin values and phototherapy rates between diabetic and normal gestational full-term newborns, providing evidence for the appropriate timing of umbilical cord clamping during delivery.

Materials And Methods

This is a prospective study. Data were systematically collected from the Department of Obstetrics and Gynecology of The Second Affiliated Hospital of Wenzhou Medical University from September 2019 to September 2020 (clinicaltrials.gov: NCT04369313). Ethics approval was obtained from our hospital (approval number: L-2019-13).

All qualified pregnant women delivered in our hospital underwent prenatal screening. Pregnant women with Rhesus negative type, abnormal liver function, hypertension, history of smoking and drug abuse, or abnormal amniotic fluid volume, preterm birth, placenta previa and other pregnancy complications were excluded. In addition, newborns also were excluded if they had a birthweight <2500g, a Apgar score ≤ 7 at 1 min or 5 min, meconium-stained amniotic fluid, neonatal malformation, or requiring neonatal resuscitation. Based on glucose test results and previous medical history, eligible subjects were divided into two groups, normal pregnant women group without any pregnancy complications and diabetic pregnant women group including pre-gestational diabetes mellitus (PGDM) and gestational diabetes mellitus (GDM).

For pregnant women without risk factors for gestational diabetes mellitus, oral glucose tolerance test (OGTT) was performed at 24-28 weeks of gestation. The diagnostic criteria for GDM were fasting plasma glucose (FPG) ≥ 5.1 mmol/L, or 1 hour (h) plasma glucose ≥ 10.0 mmol/L, or 2 h plasma glucose ≥ 8.5 mmol/L. And the FPG and HbA1C were tested at the first antenatal examination for pregnant women with risk factors for GDM. FPG ≥ 7.0 mmol/L or HbA1C $\geq 6.5\%$ was considered as PGDM. Criteria for good blood glucose control in the third trimester are the glycosylated hemoglobin (HbA1c) below 6.0% and blood glucose levels below 6.7 mmol/L at 120 minutes postprandial.

In our pre-experiments, group sample sizes of 52 and 52 achieve 90% power to detect a difference of 0.9 between the null hypothesis that both group means are 8.3 and the alternative hypothesis that the mean of group 2 is 7.4 with estimated group standard deviations of 1.4 and 1.4 and with a significance level (alpha) of 0.05000 using a two-sided two-sample t-test. Considering that unqualified cases needed to be excluded from the

trial, we increased the number of cases of every group from 52 to 70. Finally, the number of cases in diabetes pregnancy groups is 140 and that in normal pregnancy group is 140.

Consecutive pregnant women each in the group without pregnancy complications and in the group with diabetes were included according to the criteria. All the enrolled women were 18-44 years old, singleton gestation at 37 and 41 weeks plus 6 days, and signed informed consent prenatally. The women were randomly allocated to either the immediate cord clamping (ICC) or the later cord clamping (LCC) subgroup in a 1:1 ratio. Each case was identified by a unique assignment number from a computer-generated random-number list. An opaque envelope containing A or B card. According to the random sequence, the women who drew card A were assigned to the ICC group and the women who drew card B were assigned to the LCC group.

Due to the characteristics of the intervention, the mothers, the researchers performing the intervention and the assistants recording the clamping time were not masked to allocation procedure, while the physicians who measured percutaneous bilirubin value, neonatologists deciding to perform phototherapy and the statisticians conducting the data analysis were blinded to treatment.

In this study, the clamping time in the ICC group was less than 15 s after birth, while it was more than 30 s in the LCC group. Before the umbilical cord was clamped, neonates with vaginal delivery were placed on the mothers' abdomen and neonates with cesarean section were placed on the mothers' lap. Performing umbilical cord clamping on the newborn according to the original grouping and the clamping time was recorded by an assistant. The surgery was performed by obstetricians with the same surgical skills. In addition, when the mother or newborn needs resuscitation, the proper time for umbilical cord clamping is applied and the trial should be stopped immediately.

Basic demographic characteristics of the mother and newborn were recorded including mother's age, BMI, mode of delivery, gravidity, parity, gestational age, birthweight, fetal sex and Apgar scores at 1 min and 5 min. Neonatal bilirubin levels on days 2-4 and neonatal phototherapy rate were also documented. Neonatal percutaneous bilirubin levels were measured by the same physician using a unified TCB device (JM-103, Konica Minolta, Japan) at 8:00 a.m. every day. Whether the newborn undergo phototherapy was decided by the clinical neonatologist according to the recommendation of our unit.

Data were analyzed using SPSS version 20 (IBM, Armonk, NY, USA). Normally distributed variables were presented as mean $\pm$ standard deviation and analyzed by Student's t test or one-way ANOVA. Nonnormally distributed variables were expressed as median and interquartile range and analyzed by Mann-Whitney U test. Categorical variables were presented as number (%) and analyzed by Pearson's Chi-square test. The participants were stratified according to the degree of maternal glycemic control. A p value <0.05 was considered statistically significant.

Results

As shown in Figure 1, during the study period, 280 pregnant women met the inclusion criteria of this trial. Nineteen cases of postpartum neonatal transfusion syndrome, macrosomia, neonatal sepsis, congenital anal atresia, and ABO hemolysis were excluded. A final sample of 261 cases was included in this trial. In normal pregnancies, there were 67 cases in the immediate cord clamping group and 65 cases later cord clamping group. In diabetic pregnancies, there were 63 cases in the immediate cord clamping group and 66 cases later cord

clamping group. Due to the discharge of newborns, the number of remained cases used to analyze bilirubin value on day 4 in each group was 50, 43, 42 and 38, respectively.

As shown in Table 1, and Table 2, maternal and neonatal demographic characteristics such as maternal age, BMI, the mode of delivery and 1-min and 5-min Apgar scores, and birthweight were similar between the groups ($P > 0.05$).

Table 1
Demographic characteristics of mothers and neonates

	NP		DP		<i>p</i> ^a
	ICC	LCC	ICC	LCC	
	N=67	N=65	N=63	N=66	
Mothers					
Age (years)	30.6±5.2	31.0±4.6	32.1±4.4	31.1±4.9	0.276
BMI (Kg/m ²)	27.7±3.5	26.0±3.2	26.5±2.8	27.0±3.3	0.160
Blood glucose control, n (%)					
Well-controlled	-	-	56 (88.9)	60 (90.9)	0.703 ^b
Poorly-controlled	-	-	7 (11.1)	6 (9.1)	
Neonates					
Gestational age (weeks)	39.0±0.7	39.0±1.0	38.8±0.8	39.0±1.0	0.841
Delivery method, n (%)					
Vaginal delivery	16 (23.9)	14 (21.5)	23 (36.5)	22 (33.3)	0.175
Cesarean section	51 (76.1)	51 (78.5)	40 (63.5)	44 (66.7)	
Neonatal sex, n (%)					
Male	32 (47.8)	33 (50.8)	34 (54.0)	38 (57.6)	0.701
Female	35 (52.2)	32 (49.2)	29 (46.0)	28 (22.6)	
Birthweight (g)	3268.1±333.7	3328.8±324.1	3245.4±346.2	3304.2±338.6	0.498
Parity, n (%)					
primipara	21 (31.3)	18 (27.7)	15 (23.8)	22 (33.3)	0.647
multipara	46 (68.7)	47 (72.3)	48 (76.2)	44 (66.7)	
Body length (cm)	50 (49, 50)	50 (49, 50)	50 (49, 50)	50 (50, 50)	0.332
1-min Apgar score	10 (10, 10)	10 (10, 10)	10 (10, 10)	10 (10, 10)	0.547
5-min Apgar score	10 (10, 10)	10 (10, 10)	10 (10, 10)	10 (10, 10)	0.399
NP, normal pregnancy; DP, diabetic pregnancy; ICC, immediate cord clamping; LCC, later cord clamping; BMI, body mass index;					
Data were given as mean ± SD, n (%) or median (IQR)					
^a Chi-square test for categorical variables, Mann–Whitney U-test or student t-test for quantitative variables; the comparisons were conducted among four groups					
^b The comparisons were conducted between ECC and DCC from DP					

Table 2
Demographic characteristics of cases with bilirubin at day 4 of statistics

	NP		DP		<i>p</i> ^a
	ICC	LCC	ICC	LCC	
	N=50	N=43	N=42	N=38	
Mothers					
Age (years)	30.5±5.1	31.1±4.8	32.1±4.2	30.9±4.0	0.461
BMI (Kg/m ²)	27.7±3.5	25.8±3.2	26.5±3.1	27.1±3.3	0.136
Blood glucose control, n (%)					
Well-controlled	-	-	38 (90.5)	34 (89.5)	0.881 ^b
Poorly-controlled	-	-	4 (9.5)	4 (10.5)	
Neonates					
Gestational age (weeks)	38.8±0.8	39.0±0.9	38.7±0.8	38.9±1.0	0.614
Delivery method, n (%)					
Vaginal delivery	11 (22.0)	9 (20.9)	12 (28.6)	12 (31.6)	0.627
Cesarean section	39 (78.0)	34 (79.1)	30 (71.4)	26 (68.4)	
Neonatal sex, n (%)					
Male	23 (46.0)	24 (55.8)	23 (54.8)	21 (55.3)	0.744
Female	27 (54.0)	19 (44.2)	19 (45.2)	17 (44.7)	
Birthweight (g)	3280.0±359.1	3325.1±323.5	3246.4±361.9	3315.8±318.4	0.732
Parity, n (%)					
primipara	15 (30.0)	14 (32.6)	12 (28.6)	17 (44.7)	0.407
multipara	35 (70.0)	29 (67.4)	30 (71.4)	21 (55.3)	
Body length (cm)	50 (49, 50)	50 (49, 50)	50 (49, 50)	50 (50, 50)	0.651
1-min Apgar score	10 (10, 10)	10 (10, 10)	10 (10, 10)	10 (10, 10)	0.369
5-min Apgar score	10 (10, 10)	10 (10, 10)	10 (10, 10)	10 (10, 10)	0.314

In the normal pregnancy, there was no statistical difference in neonatal transcutaneous bilirubin levels on 2 to 4 days of life between the ICC group and the LCC group ($P > 0.05$) (Table 3). However, in newborns of diabetic mothers, there was a statistically significant difference in transcutaneous bilirubin levels on day 2-4 between the ICC group and the LCC group (7.65 ± 1.83 vs 8.25 ± 1.96 , $P=0.039$; 10.35 ± 2.23 vs 11.54 ± 2.56 , $P=0.002$; 11.54 ± 2.94

vs 12.83 ± 3.07 $P=0.024$), not on day 1 (4.52 ± 2.00 vs 4.43 ± 2.13 , $P=0.618$) (Table 3)). In addition, in neonates who received ICC, we found no significant difference in transcutaneous bilirubin levels on days 2-4 between the normal pregnancy and diabetic pregnancy groups ($P>0.05$). However, among newborns with a delay of cord clamping, transcutaneous bilirubin levels on the third day after birth significantly differed between the normal and diabetic pregnancy groups (10.35 ± 2.23 vs 11.54 ± 2.56 , $P=0.013$), not on other days ($P>0.05$) (Table 3).

Table 3
Comparisons of neonatal bilirubin and phototherapy between groups

	NP		DP		NP	DP	NP vs DP	
	ICC	LCC	ICC	LCC	P^a			
					ICC vs LCC	ICC vs LCC	ICC	LCC
Bilirubin at day 2 of age, <12 mg/dl	N=67	N=65	N=63	N=66				
	8.13 $\pm$ 1.50	7.96 $\pm$ 1.21	7.65 $\pm$ 1.83	8.25 $\pm$ 1.96	0.541	0.039	0.098	0.304
Bilirubin at day 3 of age, <15 mg/dl	N=67	N=65	N=63	N=66				
	10.67 $\pm$ 1.79	10.61 $\pm$ 1.86	10.35 $\pm$ 2.23	11.54 $\pm$ 2.56	0.854	0.002	0.387	0.013
Bilirubin at day 4 of age, <17 mg/dl	N=50	N=43	N=42	N=38				
	11.53 $\pm$ 2.07	11.92 $\pm$ 2.70	11.54 $\pm$ 2.94	12.83 $\pm$ 3.07	0.450	0.024	0.983	0.112
Jaundice requiring phototherapy	1 (1.5)	3 (4.6)	4 (6.3)	12 (18.2)	0.295	0.042	0.150	0.015

Regarding the comparison of phototherapy rates between groups, we found that in neonates with diabetic pregnancy, the rate of neonatal phototherapy was significantly higher in the LCC group than in the ICC group (6.3% vs 18.2%, $P=0.042$), but there were no significant differences in the normal pregnancy subjects ($P>0.05$) (Table 3). In addition, among neonates receiving LCC, the rate of phototherapy was significantly higher in the diabetic pregnancy group compared to the normal pregnancy group (4.6% vs 18.2%, $P=0.015$). In contrast, there was no significant difference in the rate of phototherapy between the diabetic pregnancy group and the normal pregnancy group among neonates receiving ICC ($P>0.05$) (Table 3).

Discussion

To our knowledge, this is the first to describe the differential effect of later umbilical cord clamping on neonatal jaundice between normal pregnancies and diabetic pregnancies. Our study found that the neonates of diabetic mothers who received LCC had the higher transcutaneous bilirubin values and neonatal phototherapy rates on days 2-4 than those who received ICC. But in the normal pregnancy group, there were no significant difference in bilirubin levels and phototherapy rates between neonates receiving LCC and those receiving ICC. In addition, we observed that compared with neonates in the normal pregnancy group, neonates in diabetic pregnancy group had higher bilirubin levels on the third postnatal day and phototherapy rates when receiving LCC, but not ICC.

Therefore, diabetic pregnant woman who receive LCC are more likely to develop neonatal jaundice than neonates with normal pregnancies.

A delay of umbilical cord clamping is a direct placental transport process that provides warm, oxygen-rich blood to the newborn, increases the baby's blood volume, improves iron storage and benefits neonatal growth[14, 15]. LCC also transports excess red blood cells into the infant, which, combined with the limited ability of the newborn liver to metabolize bilirubin, theoretically makes newborns more susceptible to jaundice. Nevertheless, many researchers have reported no significant differences in bilirubin levels and phototherapy rates in infants receiving LCC, compared with neonates receiving ICC [7, 9, 16, 17]. These studies focus on normal pregnant women or those without clear definitions. Likewise, our study found that there was no significant difference in bilirubin levels and phototherapy rates in the neonates of mothers with normal pregnancy between LCC group and ICC group. In contrast, our study showed that in diabetic mothers, bilirubin levels and phototherapy rates were significantly higher in the LCC group than in the ICC group. Similar results were found in our previous study[13]. Thus, the effects of later cord clamping on neonatal jaundice is different between normal pregnancies and diabetic pregnancies.

Gestational diabetes is characterized by insulin resistance and hyperinsulinemia in pregnancy women, which stimulates aerobic metabolism, accelerates fetal growth, and increases oxygen demand. Polycythemia and hyperbilirubinemia are thought to be compensatory mechanisms in this hypoxic state [18]. A growing number of trials have also demonstrated that diabetic pregnant woman is a risk factor for polycythemia and hyperbilirubinemia and can increase the risk of neonatal jaundice and the rate of phototherapy[11, 19–21]. Although the incidence of jaundice requiring phototherapy in term infants after later cord clamping is slightly increased[14], most studies[22, 23] and our study have confirmed that there were no significant difference in bilirubin levels and phototherapy rates in neonates of mothers without pregnancy complications between ICC group and LCC group. However, in diabetic pregnant woman, LCC was able to significantly increase neonatal bilirubin levels and phototherapy rates compared to ICC. Therefore, diabetic pregnant woman are more likely to develop neonatal jaundice compared with neonates with normal pregnancies when they received LCC. This suggests that neonates with diabetic pregnancy should be fully evaluated before performing LCC to prevent adverse consequences of elevated bilirubin.

On the other hand, compared with the normal pregnancy group, bilirubin levels on the third postnatal day and phototherapy rates of the neonates in the diabetic pregnancy group were significantly higher when neonates received LCC, not ICC. Poor maternal glycemic control versus strict glycemic control enhances the risk of neonatal complications such as neonatal hypoglycemia, erythrocytosis, jaundice, and phototherapy rates [24, 25]. In the present study, the vast majority of mothers in the diabetic pregnancy group had good glycemic control (86.6%,91.5%), so there was no difference in bilirubin level and phototherapy rate between the diabetic pregnancy group and the normal pregnancy group in neonates receiving ICC. But compared with normal pregnancy women, neonatal bilirubin level on the third postnatal day and phototherapy rate were increased in the diabetic pregnancy women after excessive placental blood transport into the neonates through LCC. So LCC can increase the risk of neonatal jaundice in diabetic pregnant woman even though the blood sugar control is good.

Our trial is the first to examine the differential effects of LCC on jaundice in newborns with diabetic pregnancy and normal full-term pregnancy. However, the main limitation of this study is that only full-term normal weight

fetuses were analyzed, which limits the generalization of this conclusion. Therefore, we will explore the effect of LCC on jaundice in preterm infants with diabetic pregnancy in the future.

To sum up, in the current study, we found the effects of later cord clamping on neonatal jaundice is different between normal pregnancies and diabetic pregnancies. There was no significant effect of LCC on neonatal bilirubin values and phototherapy rates in neonates without maternal pregnancy comorbidities, but in diabetic pregnant woman, LCC significantly increased transcutaneous bilirubin levels and phototherapy rates. Therefore, later cord clamping should be performed in healthy full-term neonates without fear of jaundice, while LCC should be cautiously performed in full-term neonates with diabetic pregnancy because of the increased risk of neonatal jaundice.

Abbreviations

FPG	Fasting plasma glucose
GDM	Gestational diabetes mellitus
ICC	Immediate cord clamping
LCC	Later cord clamping
OGTT	Oral glucose tolerance test
PGDM	Pre-gestational diabetes mellitus

Declarations

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Author contributions: S.P., Q.L.: Drafting the article or revising it critically for important intellectual content; Y.L., L.P., X.Y: Substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; Y.H.: Final approval of the version to be published.

Ethics approval: This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of Wenzhou Medical University.

Consent to participate: Informed consent was obtained from all individual participants included in the study.

Consent to publish: Not applicable.

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Figures

Figure 1

Legend not included with this version.