# ADDIS ABABA UNIVERSITY
## COLLEGE OF HEALTH SCIENCES
## SCHOOL OF PUBLIC HEALTH

**Effect of Time of Cord Clamping on Serum Bilirubin Level among Full Term Babies Born at Tikur Anbessa Specialized Hospital: A Randomized Control Trial.**

**Investigator: Mr. Biruk Hailu Tesfaye**
**Advisors: Mulugeta Betre Gebremariam (Assoc. Prof-Dr.)**
**Abiy Seifu Estifanos (MPH, BSC)**
**Asrat D. Gebremedhin (Neonatologist, MD, Assistant Prof.)**

**A Thesis Submitted to the School of Public Health, College of Health Sciences of Addis Ababa University in Partial Fulfillment of The Requirements for The Degree of Master's in Public Health-Reproductive and Family Health.**

**June, 2020**
**Addis Ababa, Ethiopia**

**Advisor's Approval Sheet**

This is to verify that the thesis entitled "*Effect of time of cord clamping on serum bilirubin level among full term babies born at Tikur Anbessa Specialized Hospital: A three arm randomized control trial.*" is submitted in partial fulfillment of the requirements for the degree of masters of public health with specialty in "Reproductive and Family Health" to the Graduate Program of the School of Public Health, College of Health Sciences of Addis Ababa University and has been carried out by Biruk Hailu under our supervision.

The student has fulfilled the thesis requirements and hence here by can submit the thesis to the school.

Name of advisors

Mulugeta Betre (Assoc. Prof-Dr.) Date: _____ Signature: _____

Abiy Seifu (BSC, MPH) Date: _____Signature: _____

**Examiners' Approval Sheet**

We, the undersigned, members of the Board of Examiners of the final open defense by Biruk Hailu Tesfaye, have read and evaluate his thesis entitled "*Effect of time of cord clamping on serum bilirubin level among full term babies born at Tikur Anbessa Specialized Hospital: A three arm randomized control trial.*". This is to verify that the thesis has been accepted in partial fulfillment of the requirements for the masters of public health degree in "Reproductive and family health"

Name of internal examiner:

_____ Date: _____ signature _____

Name of external examiner:

_____ Date: _____signature _____

## ACKNOWLEDGEMENT

Initially, I would like to give my appreciation to Mulugeta Betre (Assoc. Prof.-Dr.), Abiy Seifu and Dr. Asrat Dimtse for their unreserved advice and supports during the time taking journey of the study especially Mr. Abiy Seifu for his patience and creating each challenge as an opportunity to learn something new in my academic life.

Additionally, I would like to thank Addis Ababa University, College of Health Sciences for the financial support and for creating a comfortable environment at my area of study (Tikur Anbessa Specialized Hospital) despite the burden and sophistication in the delivery and neonatal care units. Furthermore, I would like to show how grateful I am to UNFPA for funding this study through a research project "National capacity strengthened to increase demand for and availability of family planning services, including reproductive health commodities *(project number: ETH08FPS).*

Further deepest appreciation is forwarded to the study participants, data collectors and supervisors (Especially Sr. Hawa Ali and Sr. Mulu) of the trial for being patient and highly responsible in collecting and submitting the data's properly. In addition, I would like to say thank you to my friends and family for their continuous support in different ways.

## TABLE OF CONTENTS

# LIST OF TABLES AND FIGURES

## List of Tables



## List of Figures

## ACRONYMS and ABBREVIATIONS

| | |
|---|---|
| **AAU** | Addis Ababa University |
| **ACOG** | American College of Obstetrics and Gynecology |
| **BEMONC** | Basic and Emergency Obstetrics and Newborn care |
| **DCC** | Delayed Cord Clamping |
| **ECC** | Early Cord Clamping |
| **EMwA** | Ethiopian Midwives association |
| **EPS** | Ethiopian Pediatric Society |
| **ESOG** | Ethiopian Society of Obstetrics and Gynecology |
| **EWEC** | Every Woman Every Child |
| **ICC** | Immediate Cord Clamping |
| **IRB** | Institutional Review Board |
| **LMP** | Last Day of Menstrual Period |
| **LMIC** | Low and Middle-Income Countries |
| **MOH** | Ministry of Health (Ethiopia) |
| **RBCs** | Red Blood Cells |
| **RCOG** | Royal College of Obstetrics and Gynecology |
| **SDG** | Sustainable development goal |
| **SPH** | School of Public Health |
| **SVD** | Spontaneous Vaginal Delivery |
| **TCC** | Time of Cord Clamping |
| **TORCH** | Toxoplasmosis, Other viruses, Rubella virus, Cytomegalo virus & Herpes |
| **TSB** | Total Serum Bilirubin |
| **UNICEF** | United Nations Children's Fund |
| **WHO** | World Health Organization |

## ABSTRACT

**Background:** The right timing for umbilical cord clamping has been a topic of interest for a very long time. Delayed cord clamping above one minute has linked with short and long-term nutritional and developmental benefits; otherwise delaying cord clamping associated with elevated serum bilirubin level as potential harm that could lead to jaundice requiring phototherapy at the early ages of life.

**Objective**: To evaluate the effect of the time of cord clamping on the serum bilirubin level and measure the serum bilirubin level difference between immediate and delayed cord clamping groups within 24 hours of birth from October 2019 - January 2020.

**Methods**: This study is ethically cleared three-arm, single blinded, randomized controlled study among healthy, full term neonates born in Tikur Anbessa Specialized Hospital, Ethiopia. A total of 141term babies calculated with 95% CI and 90% power as samples and enrolled in the study after screening for eligibility and have their mothers' consent. Study participants were randomly assigned across the two arms of the study; immediate cord clamping (< 1 minute and delayed cord clamping (1-3 minutes). The newborns are assessed for the outcomes at the age of 24 hour and before discharge to home using bilirubinometry and Bhutan nomogram. Basic descriptive analysis conducted to assess the maternal social, obstetric and demographic characteristics with number and percentage. Pearson Chi-square, Fisher's exact and Kruskal Wallis tests used to compare the association between maternal, fetal factors and time of cord clamping by groups. Simple and linear regression analysis used to predict serum bilirubin level from the predictor variables.

**Result:** Maternal demographic, social and obstetrics factors were not different between the groups except labour duration that had a P-value < 0.05. Time of cord clamping was not a significant predictor of total serum bilirubin levels at the age of 24 hours rather Cord blood total serum bilirubin (TSB) (coef. 0.24) and bilirubin nomogram high risk zone (Coef. 6.25) were significant predictors with P-value of 0.011 and < 0.001 respectively.

**Conclusion:** Time of umbilical cord clamping have no significant association with the total serum bilirubin level of neonates at least within 24 hours of birth.

# 1. INTRODUCTION

## 1.1 Background

According to the global estimates 386,000 babies are born every day out of which more than 90% of them are in less developed countries. From the global estimates, Ethiopia is one of the top nine countries that contribute half of the total childbirth around the world with more than 9000 births every day.(1)From the total live births in Addis Ababa (Ethiopia), around 75% of them are delivered with spontaneous vaginal delivery under the risk of facing several challenges and complications around time of birth.(2)

From the list of possible challenges around the time of birth jaundice (hyperbilirubinemia) with in the first 24 hours can be consider as one. Hyperbilirubinemia can be defined as a condition of excessive level of bilirubin (>34.2 μmol/L or 2 mg/dl) either in conjugated or unconjugated form.(3) Eighty percent of bilirubin is derived from the heme group of hemoglobin, which comes from the destruction of red blood cells in the reticuloendothelial of liver, spleen and bone marrow.(4) The destruction of red blood cells (RBCs) could occur either due to premature rupture or excessive RBCs (polycythemia) and is directly influenced by the delay in the clamping time that give extra 80-100ml blood within the first 1-3 minutes to enter the neonate circulation.(5-7)

The problem of neonatal hyperbilirubinemia globally differ due to the variability of the risk factors from region to region but it is estimated to affect about 60% of term babies out of which 10 % of them develop a severe pathologic jaundice and required phototherapy with in the first 24 hours after birth.(8, 9) The problem of jaundice and its complications are highly sensitive and needs active preparation and management modalities in order to minimize the case fatality rate (CFR).

Despite the variations in CFR between the low and middle income African countries majority of them share a CFR of between 20-60% from developing the severe neonatal jaundice and other related permanent and temporary complications are also found to be worrying.(10)In Ethiopia, the scenario related with neonatal jaundice do not defer much than the rest of Africa in that it is estimated about 13% of neonates born in hospitals develop neonatal jaundice and among them about 7% of babies develop severe related complications like bilirubin encephalopathy.(11)

In order to mitigate the complications and fatality rate of severe neonatal jaundice there are different successful interventions with the aim of preventing the development of bilirubin associated neurodevelopmental sequel, to reduce serum bilirubin levels, with minimal adverse effects.(12)

Administration of phototherapy (which employs blue wavelengths of light to alter unconjugated bilirubin in the skin) and exchange transfusion procedure (which removes partially hemolyzed and antibody-coated erythrocytes and replaces them with uncoated donor red blood cells that lack the sensitizing antigen) are found to be evidence based successful interventions in prevention and mitigation of complications from severe neonatal jaundice in the early days of postpartum.(12, 13)

The nature of the treatment options makes them difficult to be accessed easily and the complications arising from the neonatal jaundice including the case fatality rate are high in low and middle-income countries (LMICs) unless an early diagnosis and treatment of the problem initiated. Requirement of resources from the treatment option is high and optional inexpensive and easily operable interventions of educational programs for both families and health care providers could play an important role in the early diagnosis and management of neonatal jaundice without severe complications. (14)

## 1.2. Statement of the problem

A single full term newborn on average receives 24ml/kg of blood volume which is around 30% of total blood volume that the newborn will have by only implementing delayed cord clamping (DCC) to between 1-3 minutes while the newborn rests on the abdomen and parallely providing other cares related with the mother or the newborn.(3) According to existing body of literature, delayed cord clamping was reported to be beneficial in preventing neonatal hospital mortality.(15) This is mainly because of its potential to lower the risk of reduced hematocrit level at 24hours and also reducing the risk of iron deficiency at 3-6 months.(15, 16) The implementation of DCC has a good deal of support globally for the healthy outcome of neonates and mothers even though there are some arguments on adverse effects.

Despite the support from multiple studies on the implementation of delayed cord clamping for immediate and long-term benefits of both the neonate and the mother (3), small in number but emerging evidences are showing the counter effect of delaying the cord clamping, mainly attributed to the increasing neonatal risk of polycythemia and neonatal jaundice that require phototherapy.(16, 17)

Based on the findings from various randomized clinical trials and reviews, the World Health Organization (WHO) recommended that delayed umbilical cord clamping (not earlier than 1 min after birth) can improve maternal and infant health and nutrition outcomes. Additionally, one of the research areas recommended by the WHO was the investigation of the association between delayed cord clamping and hyperbilirubinemia (jaundice) on newborns.(3)Despite the recommendations, there is a paucity of data from a resource-limited setting to recommend implementation of the delayed clamping and/or proposing development of guideline/protocol for applicability.

While being under the influence of fear of unwanted counter effects on the procedure of DCC, the Many health professionals has been following a more liberal approach (according to personal communications) and that make the practice not to be applied in coherence across the country and it can prone the neonates for unnecessary related complications and loss of the expected benefits

from the easy clinical procedure of DCC.

From the expected complications arising from DCC, likelihood of developing neonatal jaundice with in the first 24 hours can be considered as the most dangerous complication and if not treated earlier could cause acute encephalopathy, kernicterus (generally are conditions that can cause temporary and permanent neurologic damage) and even the possibility of fatal outcomes cannot be neglected. The most common Factors related to the cause of pathologic jaundice within 24 hours after birth includes ABO incompatibility (a condition that happens when a mother blood type is O and her baby's blood type is A or B) , glucose 6 phosphate dehydrogenase enzyme (G6PD), polycythemia(18)and apart from the range of factors the existing evidence on DCC as cause of neonatal jaundice remains controversial between literatures. Therefore, in order to understand the influence of the practice of DCC on newborns, the contribution of a clinical trial will be eminent towards either preventing the counter effects or promoting the practice of DCC for maximum benefit possible.

### 1.3. Rationale and significance of the study

In Ethiopia, there is no consistent protocol/guideline in place for implementation of a delayed cord clamping, other than the mere incorporation of the idea in professional trainings like the Basic and Emergency Obstetrics Newborn care (BEmONC) and project activities on Helping Babies Survive (HBS) by Ethiopian Midwives Association (EMwA), Ethiopian Pediatric Society (EPS) and Ethiopian society of obstetrics and gynecologists (ESOG) for health professionals as part of an active management of third stage of labor. However, given the potential immediate and long-term benefits of implementing delayed clamping, it is imperative to investigate if delayed clamping can improve neonatal and maternal outcomes.(3) More importantly, it is important to assess the feasibility of adapting the recommendations in resource limited settings like Ethiopia where affordability for diagnosis and management of complications such as neonatal jaundice is a challenge.(19)

Therefore, this study aims to assess the potential to implement the practice of delayed cord clamping for the possible maximum beneficence and protection from harm (due to the procedure) for the newborns in Ethiopia by identifying the effect of the time of cord clamping and level of hyperbilirubinemia that requires phototherapy.

# 2. LITERATURE REVIEW

## 2.1. Historical overview of appropriate time of cord clamping

The concept of time of cord clamping resides back more than hundred years as an argumentative issue in the literatures after the initial argument of the famous English inventor/philosopher and physician Erasmus Darwin in his publication called Zoonemia.(20) Darwin states his argument as *"Another thing very injurious to the child is the tying and cutting of the navel string too soon, which should always be left till the child has not only repeatedly breathed but till all pulsation in the cord ceases. As otherwise the child is much weaker than it ought to be, a part of the blood being left in the placenta which ought to have been in the child and at the same time the placenta does not so naturally collapse, and withdraw itself from the sides of the uterus, and is not therefore removed with so much safety and certainty."*(21)After that delayed cord clamping has been a procedure of choice until the early 1900s and due to not well established reasons, the delaying of clamping time shifted to early cord clamping(i.e. 10-15 seconds after birth) around 1935. Since the year of 1935, due to different but not evidence based reasons the early cord clamping has been considered as a golden procedure in the cord management after birth.(22)

## 2.2. Why is the appropriate time of cord clamping so important*?*

### 2.2.1. Enhances placental transfusion and blood volume

Placental transfusion is literally the transfer of placental blood to the fetus during and after birth through the umbilical cord. It is a physiologic mechanism that could help anyone understand the importance of time of cord clamping in the newborns with their possible effects.(15) Newborn's benefit in terms of placental transfusion as the time of cord clamping increases from time to time. placental transfusion contribute on average 24ml/kg for the newborn (95% CI,19–32 ml) with distribution of blood volume to the infant from the placenta as a difference of corrected blood volume(CBV) from 76.3% (less than 30 seconds) to 83.9% (at 60 second) and 92.8% at 180 seconds.(23, 24) And the total volume of blood transfer increases from 25 ml in immediate clamped group to 100ml in the delayed clamped group of newborns(i.e. 75% higher among DCC groups).(15)

### 2.2.2. Improves early hematologic status and prevent anemia

Due to the variation in time of cord clamping the delayed cord clamping group has a higher level of hematocrit and hemoglobin level of58.4% & 19.5mg/dl from that of the ECC group having a 52.5% & 17.7mg/dl with in the first 24-48 hours showing a significant association.(16) In addition to that other findings also shows a significant difference in hematocrit, hemoglobin and serum ferritin level (As ferritin is the most sensitive indicator of iron status)(24) continues after 3 months(25)and even extends as long as 4-12 month.(17, 26, 27) Hence the role of delayed cord clamping is highly important in the prevention of early and mid-infancy iron deficiency anemia especially in low and middle-income countries.

### 2.2.3. Benefits for the mother

The practice of DCC for more than one minute also has well-established evidences in the literature on its significant benefits for the mother**.** Although there were arguments on the complications of post-partum hemorrhage and difficulty in administering oxytocin, Evidences suggests that the DCC procedure do not expose the women to complications related to delay in Active Management of Third Stage of Labor (AMTSL) including mean blood loss, postpartum hematocrit and post-partum hemorrhage(PPH).(28, 29)

## 2.3. Relation between time of cord clamping with neonatal Jaundice

By the year 2017, the American College of Obstetricians and Gynecologists (ACOGs) with other committees on obstetrics released a committee opinion based on existing evidence in that application of DCC on the term and preterm newborn infants has a favorable effect on the short and long-term benefits mentioned above. The committee also suggested that a slight increase in level of jaundice requiring phototherapy exist among the DCC group of infants and mechanism of monitoring and managing the jaundice problem should be in place before implementing the protocol at all levels of facility.(30)

The concept of jaundice requiring phototherapy in relation to time of cord clamping (TCC) has been a major source of argument on the inconsistencies between literatures.(31)The argument extends as long as before 1980’s(32) until recent years of 2015(25, 33-35) that delaying the time of cord clamping for more than one minute could significantly affect the serum bilirubin level of the term newborns in a way that can even requires treatment especially phototherapy.

### 2.3.1. Physiologic relation between time umbilical cord clamping and neonatal jaundice

The production of bilirubin is higher among neonates with a daily production of 8mg/kg which is about two times higher than adult production.(36) More than 80% of contribution to the production of this much higher bilirubin level is increased RBCs and polycythemia by the physiologic destruction of the cells either due to premature rupture or increased cell number.(36, 37)For the occurrence of increased RBCs in the neonate the major contribution arose from the increased blood volume that will be added to the neonatal circulation. The total amount of blood approximates to 300-350 ml for a normal child birth without an intervention and this amount of blood will increase as we delay the time of cord clamping from <1 minute to 3 minutes by 25 ml for Immediate cord clamped (ICC) up to 100-120ml for delayed cord clamped (DCC) that will approximate the total amount of blood volume to 450ml for the infants in the DCC group. These additional 100-120ml blood believed to contribute to the elevated RBCs level and indirectly to the elevated serum unconjugated bilirubin that could cause hyperbilirubinemia requiring phototherapy to be managed.(15, 38, 39)

## 2.4. What factors influence neonatal serum bilirubin level with in the first 24 hours of age?

According to different studies the risk factors for development of hyperbilirubinemia categorized broadly in to maternal, perinatal and neonatal factors. The maternal factors include ABO incompatibility, labor augmentation or induction with oxytocin, Breastfeeding, prim parity, Ethnicity/race, chronic medical illnesses like diabetes and sibling history of jaundice.(40-42)

The other directly influencing factors are associated with newborn both during perinatal and neonatal periods. These factors includes TORCH infections (a set of viral infections of fetuses including toxoplasmosis, other viruses, rubella, cytomegalovirus, herpes…etc.), elevated cord blood bilirubin level ,underweight/weight loss, Infrequent feedings, polycythemia.(40, 43)

The purpose of this study was to evaluate the effect of time of umbilical cord clamping on serum bilirubin level among full term newborns within 24 hours of birth in order to generate a knowledge towards one of the most common considered disadvantages from delaying an umbilical cord clamping of newborns for the weighing benefits.

## 2.5. Why is Delayed cord clamping important for countries like Ethiopia?

Though delaying an umbilical cord clamping time to 1 – 3 minutes is an old concept it has been through professional challenges for a significant time period and currently is being considered as an important child development intervention(3) as a component of child thriving intervention under Sustainable Development Goal 3(SDG), Every women Ever child (EWEC) global strategy of WHO from 2016 – 2030 with its early neonatal and late infancy neurodevelopment benefits.(44)

As part of the global community and World Health Organization Ethiopia has established a five-year strategy (2016 – 2020) as a component of SDG 3 and EWEC by implementing National strategy for newborn and child survival mainly focusing on the 'Survive' compartment of the EWEC global strategy with evidence-based intervention like chlorhexidine for cord care(45). The activities being implemented showed a progress on newborn and child survival(45) but needed further continuity on the health and well-being of those surviving children's by implementing evidence based developmental interventions.

## 2.6. Conceptual framework

The conceptual framework that will be used for these study is originally prepared by Nitin Pandey et.al(36) for the purpose of explaining the pathophysiology of jaundice and factors associated with Jaundice/hyperbilirubinemia. The framework in this study will try to explain the risk factor and causal relationship between delayed cord clamping and pathological jaundice within 24 hours after birth and to display the most scientifically supported factors that can cause hyperbilirubinemia with relation to time of cord clamping. The variables in different color and bold from the list of variables indicates that the exposure and outcome variables which the primary objective of the study. (*Figure 1*)

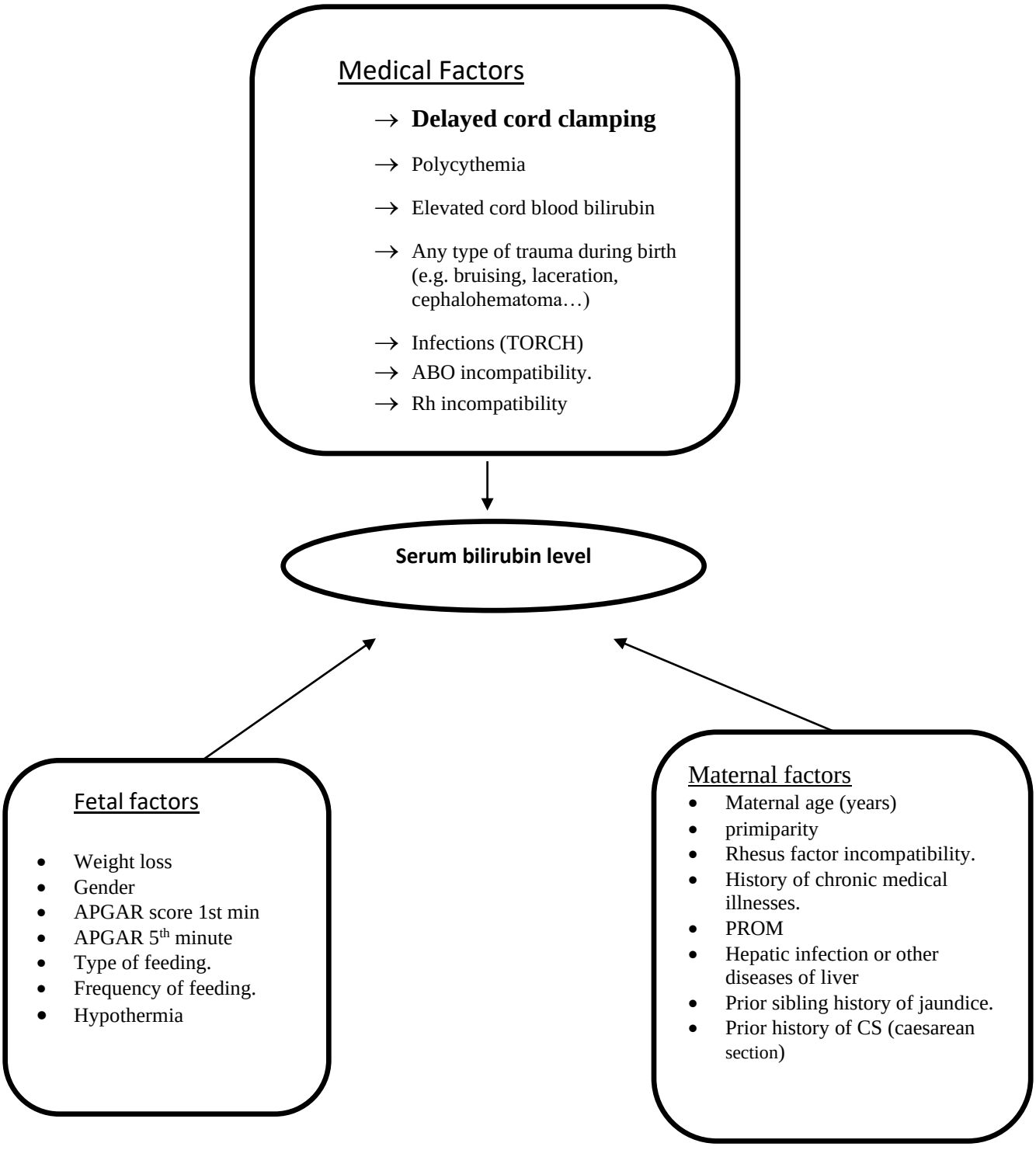


Figure 1: Conceptual relations between factors that could affect serum bilirubin level for newborns born at TASH, from October 2019 – January 2020

*(Diagram adopted from: Physiological Jaundice: Role In Oxidative Stress(p80),by Nitin Pandey et.al,2013,researchgate,vol 05(19):80.(36))*

# 3. HYPOTHESIS AND RESEARCH QUESTION

## 3.1. Research questions

- Does time of umbilical cord clamping among term babies affect their serum bilirubin level at 24 hours of age after delivery?
- Does serum bilirubin level vary when umbilical cord clamped immediately (< 1 minute) and delayed to ≥ 1-3 minutes after birth?

## 3.2. Hypothesis to be tested

- Delayed (≥ 180 seconds) umbilical cord clamping and immediate (<30 seconds) umbilical cord clamping have similar predictive effect of serum bilirubin level at 24 hours age among healthy newborns.

# 4. OBJECTIVES

## 4.1. General objective

→ To evaluate the effect of time of cord clamping on serum bilirubin level among full-term babies born from October 2019 - January 2020 at Tikur Anbessa Specialized Hospital (TASH), Addis Ababa.

## 4.2. Specific objectives

→ To determine the effect of the timing of cord clamping on serum bilirubin level among full-term babies within 24 hours of birth.

→ To measure the level of serum bilirubin among full-term babies that had their cord clamped immediately and delayed after birth.

→ To compare the proportion of full-term babies who develop hyperbilirubinemia that requires phototherapy between immediate and delayed cord clamping groups.

# 5. METHODOLOGY

## 5.1. Study area and setting

Tikur Anbessa Specialized Hospital (TASH) has a delivery unit with 11 beds for 1st stage labor follow-up and 4 couches to attend a vaginal delivery. The unit also has 20 senior (gynecology and obstetrics specialists) professionals with 93 residents of gynecology and obstetrics at different levels and 22 midwives with BSc (Bachelor of Science) and diploma levels that managed most of the vaginal deliveries according to the registry of the delivery unit in 2017/18. The neonatal department has three neonatologists and 36 nurses are currently working.

According to the registry of the delivery unit, there were 3788 spontaneous vaginal deliveries with 307 of them managed with the help of instruments in one year (From July 2017-June 2018). The current applicable procedure for the time of cord clamping is immediate as early as 15-30 seconds in the delivery unit of TASH by most of the birth attending professionals and some implement delayed cord clamping without any specific protocol on a written form.

## 5.2. Study design and period

A parallel, three arm, **equivalence randomized control trial** on healthy full-term newborns bornin TASH from the period of October 2019 to January 2020.

## 5.3. Population

### 5.3.1. Source population

All single full-term newborns (37 and above weeks of gestation) with spontaneous vaginal deliveries (SVD) without complications in between October 2019 - January 2020 in Addis Ababa.

### 5.3.2. Study population

All single full-term newborns born with SVD and no gross complications in TASH from October 2019 - January 2020.

### 5.3.3. Study unit

Selected full-term newborn born with SVD and without gross complication of any kind in TASH, assigned randomly to any of the groups from October 2019 - January 2020.

### 5.3.4. Eligibility criteria

All full-term babies given birth in TASH in between October 2019 - January 2020and have no gross complications.

### 5.6.1. Inclusion criteria

Those newborns with gestational age of ≥ 37 weeks as diagnosed with first day of last menstrual period (LMP) or ultrasound and that had no gross complications related with the pregnancy and delivery like mal-presentation, fetal distress, and congenital malformations were included in the study.

The gestational age, and fetal related information's to include in the study were referenced from asking the mothers about their ANC history and cross-referencing their medical record chart (health provider note at each visit) and the fetal distress was also referenced from the labour following partograph and consecutive fetal heart rate monitoring's.

### 5.6.2. Exclusion criteria

Those mothers with any medical or obstetric complications like hepatitis, HIV/AIDS, hypertensive disorders of pregnancy, diabetes and severe anemia were excluded from the study initially by asking them for their history and newborns with known congenital malformation during pregnancy or delivery were also excluded. Furthermore, newborns with confirmed multiple pregnancy and mal-presentation and those required immediate resuscitation after birth were also not eligible in this study as referenced from the maternal ANC records (health providers note and ultrasound results).

## 5.4. Intervention implementation modalities

After acquiring ethical approval from the Institutional Review Board (IRB) of Addis Ababa University/School of Public Health (AAU/SPH), the eligible participants enrolled in three groups for each delivery after an informed consent obtained from the mothers: the groups (ARMs)were (I)Immediate cord clamping (ICC) within 60 seconds of birth, and (ii) delayed between one to three minutes clamping (DCC) after birth. Participants who have agreed to participate in the study were randomly assigned using a table of random numbers. Group of allocation of the participants revealed only for the birth attendant (i.e. participants will not know their assignment groups) after the participant has reached at least a cervical dilatation of eight centimeters by opening an initially prepared envelope by the supervisors.

Following the random assignment of eligible participants, the umbilical cord was managed by the birth attendant according to the newborn's group of assignment.

1. The immediate clamping group received an intervention of clamping and cutting the umbilical cord immediately after expulsion/delivery. This group has received the cord management immediately (< 60 seconds/one minute) after expulsion of body of the fetus with a follow up by the supervisor.

2. The delayed group received a similar intervention except that the clamping of the cord time was at/greater than 60 seconds (1-3 minutes) and the baby was well wrapped with a towel and the minute monitored by the supervisor and he/she did communicate the attendant when it is time for the intervention. This group was closely monitored for the specific seconds to clamp the cord by the assigned supervisor.

An initial 1mililiter of cord blood was drawn using a syringe before clamping of the cord and after delivery for every participant including and transferred immediately to the laboratory unit (using anti-coagulant test tube) for the assessment of total cord blood serum bilirubin (TSB), red blood cells (RBCs) count and blood group of the neonate. The rest of the intra and post-partum care proceeded as the standard management of childbirth. Careful observation and follow-up of the mother and newborns provided by the midwives on duty as per the existing protocol for 24 hours.

### 5.5. Variables and measurements

→ The primary outcome variable was the serum bilirubin level of the newborn which was measured using laboratory bilirubinometry (*bilirubin assay kit MET-5010*) and by labeling on a Bhutan Nomogram (i.e. a bilirubin risk zone predictor) *(Annex 5)* and the secondary outcome is related hyperbilirubinemia that requires phototherapy. Likewise, RBC count was measured using a complete blood count (CBC) hematology analyzer (*Sysmex XP-400*) after 24 hours of birth and discharge of the neonate to home for all the three arms. The predictive factors or variables can be broadly classified as maternal related, fetal related and medical associated factors. The most significant maternal related variables include maternal age, primiparity, blood group & Rh and prior sibling history of jaundice. In addition to that fetal related common factors that include birth weight,

gestational age, DCC, blood group & Rh, TORCH (Toxoplasmosis, Other viruses, Rubella virus, Cytomegalo virus & Herpes) Infections and cord blood bilirubin & RBCs level could highly influence the level of jaundice directly or indirectly.

## 5.6 Sample Size Estimation and Sampling Technique

### 5.6.3. Sample size determination

Estimation of sample size for this study has drawn from the study experience of Judith S. Mercer et.al (16) using the power and sample size calculator from STATA 14 software. The sample size estimation used a significant level of p-value <0.05with α=5% and a power of 90% using a difference between proportion of newborns with jaundice that required phototherapy (11%) among the delayed cord clamped groups in comparison to early cord clamped group of newborns with proportion of < 1% (sample size using two independent proportions).The calculation generates a total sample size of 141 including 10% non-response rate and hence the study had three independent groups with 47 samples per single group.

| | |
|---|---|
| Confidence level | 95% |
| Power | 90% |
| Significance level (P-value) | < 0.05 |
| Percentage of newborns required phototherapy | |
| 3-minutes (180seconds) delayed group | 11% |
| Immediate (15 – 30 seconds) group | 1% |

$$\mathbf{n = (Z_{\alpha/2}+Z_{\beta})2 * (p1(1-p1) + p2(1-p2)) / (p1-p2)2}$$

where $Z_{\alpha/2}$ is the critical value of the Normal distribution at $\alpha/2$, $Z_{\beta}$ is the critical value of the Normal distribution at $\beta$ and p1 and p2 are the expected sample proportions of the two groups.

### 5.6.4. Sampling technique

The study area (TASH) was selected purposively for the ease of data collection in terms of the higher number of cases per month and to use the intended data collection time effectively. Selection of the study participants was done using the inclusion criteria and the mothers received an informed consent

and once they agreed the neonates were randomly assigned in to either of the three comparative groups by using the pre-determined codes generated by Excel random generator and with opening the sealed envelopes immediately before delivery. The recruitment of participants continued until it reaches the required sample size. (*Figure 02*)

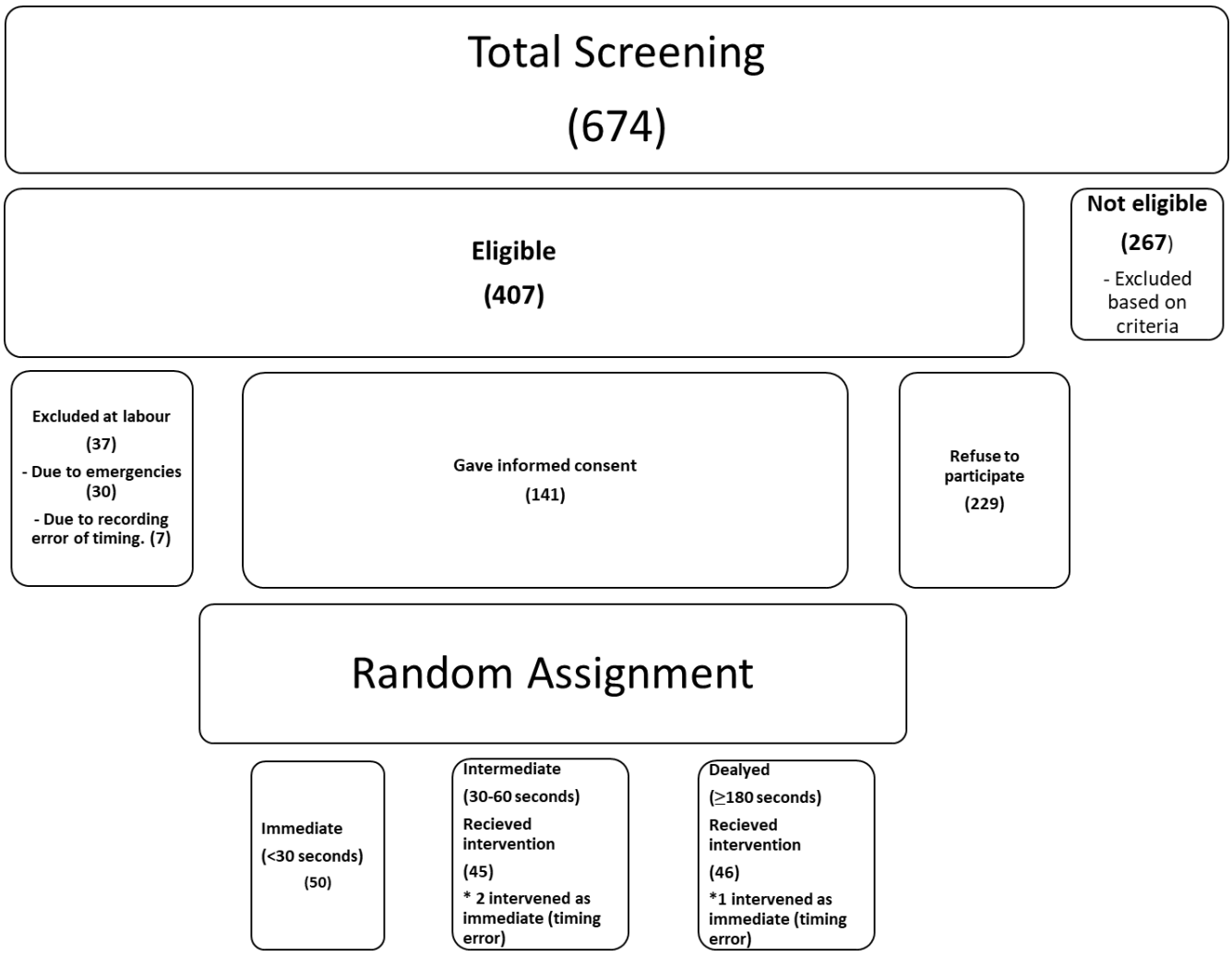


Figure 2: Sampling technique of study participants from mothers came for child birth at TASH from October 2019 – January 2020.

### 5.7. Data collection procedure

The data collection had three group of participants with a two phase data collection; 1st around perinatal phase for both maternal and fetal characteristics and other data that can be collected on the time, 2ndphase starts at the age of 24 hour after delivery; the hospital stay hours are based on the WHO recommendation of a minimum of 24 hours' post-partum for normal vaginal delivery and indicated as part of the informed consent. Blood samples for the primary outcome were collected from veins and were transported to the laboratory with anti-coagulated test tube by the supervisor within 30 minutes of the sample collection. The source of sociodemographic and obstetric data was the medical record chart of both the mother and the newborn. The rest of the data was recorded from the direct interview of the mother and laboratory findings. The data collection was done by Bachelor/Masters of Science (BSc/MSc) Midwives and/or Medical interns/residents who were attending majority of the deliveries in the hospital. The data collectors first discussed about the procedure and explained all the risk and benefits to the eligible participant and/or companion later in the first stage of labor (after cervical dilatation reaches 8cm) and receive an informed consent either from her or companion as soon as they understood, agreed to participate and before the second stage labor ensued or upon preparing the mother for delivery. As soon as the mother is ready for delivery, the randomly assigned sealed-envelope was opened by the supervisor for the birth attendant to distinguish the time of cord clamping that he/she will have intervened and the supervisor could monitor the clock with digital second counter and could inform when the intended minute of cord clamping has reached to the birth attendant. Additionally, the supervisor had the obligations in following the fidelity of the trial protocol *(Annex 4)* with a preexisting and printed checklist as part of the standard operating procedure (SOP) *(Annex3)*. After expulsion of the newborn, the birth attendant needed to collect 1mililiter (ml) cord blood and handed to the assistant after separating the cord irrespective of the group assignment. The collected blood samples were stored in a test tube and transported to the laboratory within 30 minutes after collection and the results were recorded on the newborn chart as per the usual trend by the attendant later.

### 5.8. Data collection tools and materials

The detailed maternal and perinatal characteristics were recorded based on pre-designed questionnaire, notably for the maternal details such as age, parity, mode of delivery and medical conditions from available records ***(Annex2***). Likewise, the perinatal details including gestational age, Apgar score at 1$^{st}$ and 5$^{th}$ minute, fetal presentation were also recorded ***(Annex2.)***

The recording of the outcome and confounding variables were considered by using an adapted standardized questionnaire that had been used in other similar trials (16, 42) with some modification of the questions and variables to suit the context.***(Annex 2)***

### 5.9. Randomization and masking

The randomization technique started with random assignment of participants in to blocks and the order of the sequential assignment for the three groups of participants were done using random sequence generator from Excel 2016 program available on Office 2016 package software (Microsoft Corporation).

The allocation of the participant mothers was done after obtaining their informed consent earlier in the late first stage labor (i.e.≥8cm cervical dilatation). The principal investigator (PI) had prepared the allocation codes with sealed, opaque and identically colored envelopes initially at the beginning of the study with ease of accessibility by the supervisor only. Supervisors were not involved in the screening of participants and they could reveal the envelope to the birth attendant only upon the mother enters the second stage of labor or readied for child birth proper.

### 5.10. Blinding

The data collectors were not blinded due to the characteristics of the study but the participants were not told the intervention they were receiving and that make the intervention single blinded.

### 5.11. Operational definitions

- **Healthy full-term newborn:** a newborn baby born between the age of 37- 42 with a respiratory rate of <60 breaths/minute, a pulse rate of 100-160 bpm, a temperature of 36.5-37.4 °C and without either one of gross complications like congenital malformation, severe asphyxia requiring resuscitation, twin/multiple pregnancy, umbilical cord malformations/snap…etc.
- **Immediate cord clamping** – it is a clamping and cutting of the umbilical cord immediately (i.e. <60 seconds/one minute) after birth.
- **Delayed cord clamping** – is a clamping and cutting of umbilical cord at or above 3 minutes (≥180 seconds) after birth.
- **Hyperbilirubinemia –** Is when the total serum bilirubin level rise above the 95$^{th}$percentile for age during the first week of life.(46, 47)
- **Pathologic hyperbilirubinemia** – Appearance of jaundice within 24 hours due to an increase in serum bilirubin level beyond 5mg/dl/day.
- **Physiologic hyperbilirubinemia**– Attributable to physiologic immaturity and appears between 24 -72 hours after birth and was differentiated with the measurement of the indirect bilirubin test and the value under normal status must not exceed the 18mg/dl limit.

### 5.12. Data quality assurance

The data quality management followed a series of activities at every level of the data management and started initially by random assignment of participant and masking as much as possible in order to minimize selection biases and maintain a fair allocation of the participants. The measurement of the investigations was handled by using standard laboratory techniques and the outcome assessment was done through very well experienced health professionals at the hospital with known ethical care provision. For the data collection tool, translation in to Amharic and pretest were done on 10% of the total samples before starting the data collection process. The data collectors and supervisors were also received an extensive training on the standard operating procedures (SOPs) and procedure references were posted visibly in the delivery unit/at the hand of the supervisors. The supervisors supported and supervised the data collectors on a daily basis through all the three shifts of the 24-hour service

provision. The principal investigator (PI) had to overlook and supervise the data collections on the site every 3-4 days and every day through telephone-based regular contacts.

### 5.13. Data management and analysis

After the data was collected, the completeness and consistency of the questionnaire was checked. Then both the questionnaires and the variables were coded. After this, Epidata manager version 4.4.2.1 used to enter and clean the collected data. Intention to treat analysis was used to analyze data after the completion of data collection process. The analysis was done using STATA 14 software.

Descriptive analysis was used for baseline characteristics of the participants, and was reported using the mean and standard deviation (SD) for discrete continuous data type while frequencies and percentages used for categorical data. Chi-square analysis (Pearson chi-square test) was used to analyze the association between time of cord clamping and hyperbilirubinemia requiring phototherapy. Fisher's exact test was used for observations counting less than five for all groups and Kruskal Wallis rank test used to determine a significant difference between grouped independent variables and continuous dependent variables. Simple linear regression was used to predict the effect of time of cord clamping on serum bilirubin level and also multiple linear regression was implemented to control other predictive factors in a single model. The findings were considered as significant if $P<0.05$ obtained at 95% confidence interval (CI).

### 5.14. Ethical considerations

The study proceeded after receiving an ethical clearance from School of Public Health and school of medicine (Department of gynecology) and College of Health Science, Addis Ababa University. An approval letter was submitted to College of Health Sciences and followed by the hospital's Gynecology and Obstetrics Department in order to get a professional and administrative permission to conduct the study followed by official registration of the trial on **http://pactr.samrc.ac.za/** website (an international registration of clinical trials website).

The study participants were only enrolled after receiving an informed consent by mothers of the newborns with extensive explanation of the possible ethical issues including the risk, benefits, autonomy and fairness in allocation of the sampling procedure.

The autonomy of the mother was respected by ensuring extensive explanation of the trial related matters and reaching at a well-informed decision. The right to have a query at any time of the study period through the contact addresses of the principal investigator and advisors was made easily accessible on the consent form. And equally importantly the mother also had full right to discontinue from the study at any point in time with in the data collection period.

The mothers didn't receive any direct benefit from participation but the participation was believed to highly benefit the newborn baby and future newborns of Ethiopia including their future newborns (if planned) by preventing them from iron deficiency anemia and neuro developmental deficit until the first 6 months and further up to one year.

In the event that this study could have prone to the newborns' complication of an elevated serum bilirubin level with in the first 24 hours and so considered if the level reached the treatment/phototherapy requiring stage, they were planned to be linked to the neonatology department immediately for appropriate management.

In considering the fairness of the trial to all eligible populations the allocation of the participants was done in a careful randomization technique and the participants were blinded about the group they were assigned to and that adequate training was provided to the data collectors and the supervisors on the careful handling of information.

# 6. RESULTS

## 6.1 Basic characteristics of participants

Table 1 describes the basic demographic, social and obstetric characteristics of participant mothers. As categorized under the table, about 102 (72%) of the participants were between the age of 20-34 years and 29 (20%) of them were under the age of 20 years followed by 10 (7%) above the age of 35 years. And 107 (75.88%) of the participants have had an antenatal visit of more than four times and the rest 34 (24.46%) had an antenatal visit below the standard of four times until term. From the total of 141 laboring mothers 127 (90.07%) and 14 (9.93%) had a normal and prolonged labor respectively

Table 1: Demographic and obstetric Summary characteristics of mothers gave birth at TASH, from October 2019 – January 2020

| | **Frequency** | **Percentage** |
|---|---|---|
| **Age of the mother** | | |
| <20 | 29 | 20.57 |
| 20-34 | 102 | 72.34 |
| >=35 | 10 | 7.09 |
| **Gravidity** | | |
| Primiparous | 58 | 41.13 |
| Multiparous | 83 | 58.87 |
| **ANC visits** | | |
| <4 times | 34 | 24.12 |
| >=4 times | 107 | 75.88 |
| **Labor Duration** | | |
| Normal | 127 | 90.07 |
| Prolonged | 14 | 9.93 |
| **Labor onset** | | |
| Spontaneous | 120 | 85.11 |
| Induced | 21 | 14.89 |
| **PROM** | | |
| Yes | 19 | 13.48 |
| No | 122 | 86.52 |
| **Caesarean section history** | | |
| Yes | 15 | 10.64 |
| No | 126 | 89.36 |

### 6.2. Association between maternal and obstetric factors with time of umbilical cord clamping

The association between maternal, obstetrics factors and time of umbilical cord clamping gets described in table 02. Most of the factors does not show a significant association among the categories of time of cord clamping except duration of labour which has a prolonged a P-value of < 0.05 between the immediate clamping and delayed clamping groups.

Table 2: Association between maternal and obstetric factors with time of umbilical cord clamping for mothers who gave birth at TASH from October 2019 – January 2020.

| **Variables** | **Immediate clamping (<1minute) (n=95)** | **Delayed (>=1 min) group (n=46)** | **P-value** |
|---|---|---|---|
| **Maternal Age – n (%)** | | | *0.194* |
| **<20** | 21 (22.1) | 8 (17.4) | |
| **20-34** | 65 (68.4) | 37 (80.4) | |
| **>=35** | 9 (9.5) | 1 (2.2) | |
| **Gravidity – n (%)** | | | *0.483* |
| **Primiparous** | 41 (43.15) | 17 (37) | |
| **Multiparous** | 54 (56.85) | 25 (63) | |
| **Chronic Medical illness history – n (%)** | | | *0.80* |
| **Yes** | 11 (11.6) | 6 (13) | |
| **No** | 84 (88.4) | 40 (87) | |
| **PROM – n (%)** | | | *0.52* |
| **Yes** | 14 (14.8) | 5 (10.8) | |
| **No** | 81 (85.2) | 41 (89.2) | |
| **Sibling history of jaundice – n (%)** | | | *0.24[fe]* |
| **Yes** | 1 (1) | 2 (4.3) | |
| **No** | 94 (99) | 44 (95.7) | |
| **CS history – n (%)** | | | *0.91* |
| **Yes** | 10 (10.5) | 5 (10.9) | |
| **No** | 85(89.5) | 41 (89.1) | |
| **Labour Duration – n (%)** | | | *0.032* |
| **Normal** | 82 (86.3) | 45 (97.8) | |
| **Prolonged** | 13(13.7) | 1 (2.2) | |
| **Labour onset – n (%)** | | | *0.35* |
| **Spontaneous** | 79(83.15) | 41 (89.1) | |
| **Induced** | 16(16.5) | 5 (10.9) | |
| **ABO incompatibility – n (%)** | | | *0.795* |
| **Yes** | 9 (9.5) | 5 (10.9) | |
| **No** | 86 (90.5) | 41 (89.1) | |
| **Rh incompatibility – n (%)** | | | *0.636* |
| **Yes** | 6 (6.3) | 2 (4.3) | |
| **No** | 89 (93.7) | 44 (95.7) | |
| **PPH – n (%)** | | | *0.354* |
| **Yes** | 3(3.15) | 3 (6.5) | |

| | | | |
|---|---|---|---|
| **No** | 92 (96.85) | 43 (93.5) | |
| ***fe - fisher's exact test*** | | | |

### 6.3. Newborn factors association with time of cord clamping (TCC)

Table 03 indicates whether factors categorized as newborn source have an association with time of cord clamping and compared between categories of immediate (<1 minute.), and Delayed (≥180sec.) and indicated gender ratio between male and female newborns were proportional between those categories. As indicated in the table, majority (94.6%) of the newborns were fed with breastmilk only but found to have no any association with their time of cord clamping, and 1$^{st}$ and 5$^{th}$ minute APGAR score of less than seven were 23.1 % and 3.2% respectively but had no significant association with the umbilical cord clamping time. Additionally, majority of the newborns (97.8%) were in low risk zone of the bilirubin nomogram with in the first 24 hours of birth and only three (2.1%) newborns required phototherapy in the later 48-72 hours of their birth but not significantly related with their time of cord clamping. The mean weight during birth was 3251gm (±60) and 3247gm (±67) respectively for immediate and Delayed (≥180sec.) of umbilical cord clamping time; similarly, the average temperature, cord blood RBC and cord blood TSB indicated in the table between groups but had no significant association with the newborns umbilical cord clamping time.

Table 3: Association of neonatal factors with time of umbilical cord clamping time of newborns born at TASH from a period of October 2019 – January 2020.

| **Variables** | **Immediate clamping (<1minute) (n=95)** | **Delayed (>=1 min) group (n=46)** | **P-value** |
|---|---|---|---|
| **Gender – n(%)** | | | *0.758* |
| Male | 49 (57.6) | 25 (54%) | |
| Female | 46 (42.4) | 21 (46%) | |
| **Type of Feeding – n(%)** | | | *0.078 fe* |
| Breastmilk only | 88 (94.6%) | 45 (98%) | |
| Formula feeding | 3 (3.2) | 0 | |
| Mixed Feeding | 2 (2.2) | 1 (2%) | |
| **1st minute APGAR Score – n(%)** | | | *0.08* |
| <7 | 22 (23.1) | 5 (11%) | |
| >=7 | 73 (76.9) | 41 (89%) | |
| **5th minute APGAR Score – n(%)** | | | *1.00 fe* |
| <7 | 3 (3.2) | 1 (2%) | |
| >=7 | 91 (96.8) | 45 (98%) | |
| **Bilirubin nomogram risk zone – n(%)** | | | *0.697 fe* |
| Low risk zone | 91(97.8) | 44 (96%) | |
| Low-intermediate risk zone | 1 (1.1) | 1 (2%) | |
| High risk zone | 1 (1.1) | 1 (2%) | |
| **Phototherapy** | | | *1.00 fe* |
| Yes | 2(2.1) | 1 (2%) | |
| No | 93 (97.9) | 45 (98%) | |
| **Fetal infections** | | | *0.719* |
| Yes | 8 (8.4) | 3 (7%) | |
| No | 87 (91.6) | 42 (93%) | |
| **Weight (gm) -** Mean (SD) | 3261(60) | 3247(67) | *0.935 (kw)* |
| **Temperature -** Mean (SD) | 36.14(0.08) | 36.22(0.08) | *0.347 (kw)* |
| **Cord Blood RBC -** Mean (SD) | $4.57x10^6$ (0.07) | $4.61x10^6$ (0.07) | *0.489 (kw)* |
| **Cord Blood TSB -** Mean (SD) | 1.52 (0.1) | 1.52 (0.16) | *0.57 (kw)* |
| *fe - fisher's exact test, kw – Kruskal Wallis test* | | | |

## 6.4. Effect of time of cord clamping on total serum bilirubin level at 24 hours after birth

A simple linear regression of total serum bilirubin amount with umbilical cord clamping time as indicated in Table 4 shows a negative relationship between the two variables but found as non-significant with a P-value = 0.19.Table 05 shows rather a multiple regression of other pertinent factors like bilirubin nomogram high risk zone and cord blood bilirubin has a P-value of < 0.001 and < 0.05 indicating a significant predictive relationship with serum bilirubin levels within 24 hours of birth and

delaying cord clamping time to three minute shows no any significant relation with the total serum bilirubin levels within 24 hours of birth.

Table 4: Simple regression of the total serum bilirubin level by time of cord clamping of newborns born at TASH from a period of October 2019 – January 2020.

| TSB | Coef. | Std. Err. | t | P-value | $R^2$ |
|---|---|---|---|---|---|
| **Time of cord clamping** | -0.069 | 0.099 | -0.69 | 0.489 | ***0.012*** |
| **_const** | 1.911158 | .2428028 | 7.87 | 0.000 | |

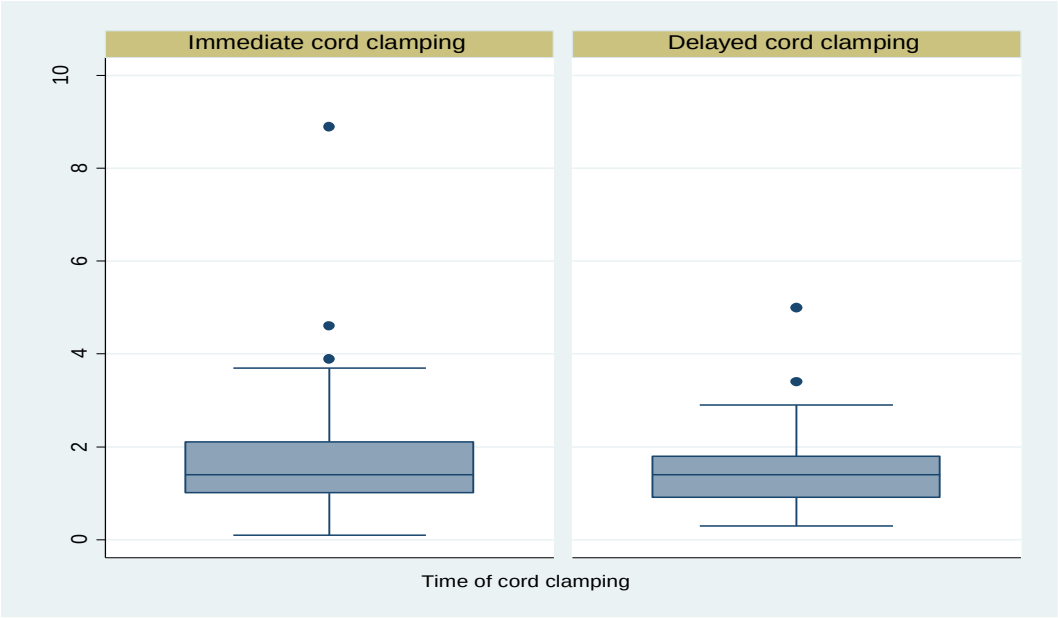


Figure 3: Relationship between time of cord clamping and total serum bilirubin level at the age of 24 hours of birth among neonates born at TASH from a period of October 2019 – January 2020.

Table 5: Multiple regressions of total serum bilirubin level as affected by different predictors within 24 hours of birth among neonates born at TASH from a period of October 2019 – January 2020.

| **Source** | **SS** | **df** | **MS** | **Number of obs** | **=136** |
|---|---|---|---|---|---|
| | | | | F | = 13.23 |
| **Model** | 79.2577379 | 9 | 8.80641533 | Prob > F | = 0.0000 |
| **Residual** | 83.8826621 | 126 | .665735413 | R-squared | = 0.4858 |
| **Total** | 163.1404 | 135 | 1.20844741 | Adj R-squared | = 0.4491 |
| | | | | Root MSE | = 0.81593 |

| TSB | coeff. | St. Err. | t | P>t | Beta |
|---|---|---|---|---|---|
| | | | | | |
| Time of cord clamping | -0.07 | 0.09 | -0.87 | 0.385 | -0.057 |
| Cord blood TSB | 0.18 | 0.09 | 2.08 | **0.04** | 0.149 |
| Cord blood RBC | -0.15 | 0.14 | -1.03 | 0.304 | -0.067 |
| Type of feeding | -0.02 | 0.22 | -0.07 | 0.942 | -0.005 |
| Newborn Weight | -0.00005 | 0.0002 | -0.3 | 0.767 | -0.019 |
| Labour duration | 0.0005 | 0.01 | 0.05 | 0.959 | 0.004 |
| ABO - incompatibility | 0.09 | 0.24 | 0.37 | 0.713 | 0.023 |
| Rh - incompatibility | -0.33 | 0.30 | -1.1 | 0.274 | -0.072 |
| Bilirubin_nomogram_risk_zone | 2.33 | 0.27 | 8.54 | **0.000** | 0.601 |
| _cons | | -0.062535 | 0.9695795 | -0.06 | 0.949 |

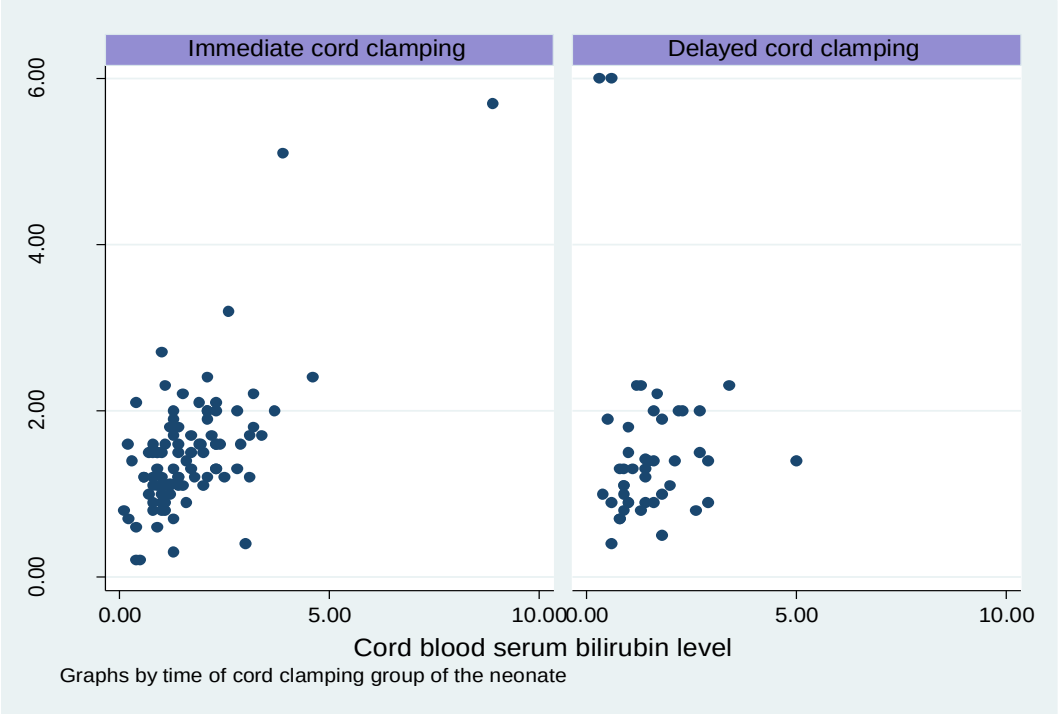


Figure 4: Predictive relationship between cord blood TSB and TSB at the age of 24 hours of birth

among neonates born at TASH from a period of October 2019 – January 2020.

### 6.5. Mean total serum bilirubin (TSB) level and difference of selected fetal factors

A mean difference between 3-minute (delayed) cord clamping and immediate cord clamping groups was -0.293 in that it was found to be higher among the immediate group. Similarly, Rh incompatibility, 1st and 5th minute APGAR score has a negative mean difference that has no significance between the groups. On the other hand, fetal infections (TORCH) and ABO incompatibility had a small positive mean difference (0.26 and 0.03 respectively) with no significance *(table 06)*.

Table 6: Mean difference between selected fetal factors within 24 hours of birth among neonates born at TASH from a period of October 2019 – January 2020.

| | Total serum bilirubin (mg/dl) Mean (SD) | Mean Difference | CI (95%) |
|---|---|---|---|
| **Time of cord clamping** | | | |
| Immediate | 1.662 (0.12) | -0.276 | 1.403346-2.231654 |
| Delayed | 1.524 (0.13) | | 1.255437-1.793452 |
| **Fetal infections (TORCH)** | | | |
| Yes | 1.87 (0.34) | 0.266 | 1.19708 - 2.54292 |
| No | 1.60 (0.09) | | 1.410165 - 1.796764 |
| **ABO incompatibility** | | | |
| Yes | 1.652 (0.22) | 0.038 | 1.1991 – 2.105 |
| No | 1.613 (0.1) | | 1.414498 – 1.813082 |
| **Rh incompatibility** | | | |
| Yes | 1.15 (018) | -0.496 | 0.785 – 1.514 |
| No | 1.646 (0.09) | | 1.452 – 1.84 |
| **1st minute Apgar score** | | | |
| **<7** | 1.425 (0.51) | -0.198 | .4154928 - 2.434507 |
| **>=7** | 1.623433 (0.09) | | 1.435365 - 1.811501 |
| **5th minute Apgar score** | | | |
| **<7** | 0.95 (0.05) | -0.672 | 0.851122 - 1.048878 |
| **>=7** | 1.622 (0.09) | | 1.434731 - 1.810306 |

# 7. DISCUSSION

This study tried to test a hypothesis that assumed delayed and immediate umbilical cord clamping have a similar predictive effect on Total serum bilirubin (TSB) level of healthy full term newborns at the 24 hours of age after birth and found that there was no significant difference between time of umbilical cord clamping and total serum bilirubin levels as measured at the age of 24 hours after birth. A group of literatures indicated that there is another contrary concept of a significant effect showed on serum bilirubin level associated with time of cord clamping is in terms of exposing the newborns to jaundice and its complication(25, 33-35); our study indicated a non-significant difference between the cord clamping time and serum bilirubin level measured and unlike the above group of literatures and it even showed a higher median TSB level *(Figure 3)* among those newborns received an immediate cord clamping less than 30 seconds.

Delaying umbilical cord clamping to 1-3 minutes is a recommended procedure by different literatures and World health organization (WHO) without compromising other equivalent routine neonatal care activities (3, 48). It can enhance the hemoglobin level of newborns from the first hours of birth to months and that can help in development of the neonatal and long-term infant outcomes. Another emerging group of literature are arguing that the procedure has a higher risk of exposure for the newborns to unwanted effects related with elevated serum bilirubin.(16, 17, 48) Supporting the DCC recommendations, our study findings observed delaying an umbilical cord clamping time is an easy and uncomplicated procedure that can be done by the birth attending professionals easily and it can substantially benefit newborns in low and middle-income countries like Ethiopia where nutrient deficiency is a common tragedy.

The appropriate time of cord clamping varies across the existing literatures and most of the studies have used variable time to clamp the umbilical cord and categorize it as early or delayed.(16, 17, 49) Our study considered categorizing the study participants in to immediate group (<30seconds) by considering the time required to manage the birth with complete expulsion of the body and for the delayed (1 – 3minute) tried to categorize them as delayed from one to three minute based on previous literatures and WHO recommendation for time of cord clamping for newborns(19, 42) for the purpose of contribution in un-defined time of delayed cord clamping among existing literatures.

A study conducted on effects of delayed cord clamping on residual placental blood volume, Hemoglobin and Bilirubin Levels in Term Infants indicated by its finding that there was a significant (P-value <0.05) increase on the number of neonates required phototherapy due to delaying the umbilical cord clamping time to 3 minutes (16); In contrary to these study, our finding indicated that there were three newborns that developed jaundice and required phototherapy as one in each arm and had no significant relationship *(table 03)* with delaying the clamping time to three minutes. This may be attributed to the fact that in our study serum bilirubin level measured within 24 hours after birth and that can minimize the chance of capturing the late (48-72 hours) elevations in serum bilirubin levels on health neonates after birth.

Elevated cord blood RBC level or polycythemia was found to be one significant factor in elevation of total serum bilirubin around the early hours of life (17, 50); from our study the cord blood RBC count not found to be significantly different or elevated above normal range between each groups *(table 03)* and it implied that delaying an umbilical cord clamping time couldn't be considered as a harming practice for the newborns. On the contrary, our study shows that cord blood TSB is a significant predictor of serum bilirubin level after 24 hours of birth.

In our study, Bilirubin nomogram high risk zone was found to be a very significant *(Reg coef. = 6.25, P-value < 0.001)* predictor of total serum bilirubin level within 24 hours of birth unlike a study by *Judith et.al(16)*that had an insignificant P-value of *0.95;* This discrepancy could be due to the chance that our study had only one participant in the high-risk zone but using bilirubin nomogram believed to be a golden standard to clearly predict, monitor and manage elevated bilirubin level at the early hours of the newborn before jaundice and further complications developed.(51, 52)

The mean serum bilirubin difference between delayed and immediate cord clamping groups was expected to be 2mg/dl as a study by *Judith et.al* (16), from our results we found a difference of -0.293 mg/dl indicating a higher mean value among the immediate groups and this might be attributable to the time of measurement in the previous study (16) was at 72 hours and ours measured at 24$^{th}$ hour after delivery of the newborn.

Similar to prior studies(37, 40-42)factors like Postpartum hemorrhage, ABO incompatibility and Rh incompatibility showed no significant association with the time of cord clamping with a P-value of *0.59, 0.85, 0.52)*with an implication of these factors shouldn't be sources of bias for avoiding the delaying the time of cord clamping for healthy neonatal and infant outcomes.

Fear of unwanted side effects like hyperbilirubinemia, habitual practices related with age of birth attendants, type and level of profession and number of births in the facility are found to be the most common barriers for the successful implementation of delaying umbilical cord clamping in the global community of maternal and child birth service providers.(53-55) From our study results the issues related with side effects of hyperbilirubinemia found to be insignificant and the study considered the practice of delayed cord clamping is a safe practice in terms of unwanted bilirubin related side effects.

# 8. STRENGTH AND LIMITATIONS OF THE STUDY

## 8.1. Strengths of the study

The primary strength of the study relies on designing the study in a more acceptable, controlled and randomized way using a Randomized allocation of all potential variables for generating a strong and straightforward evidence towards effect of time of cord clamping (especially delayed) on serum bilirubin level.

In addition to the design of choice, implementing the proper protocols of an RCT in such a resource limited and busy study area (TASH) can be considered a strong aspect of this study starting from providing an intensive training to contributors, block randomization and careful implementation of the study protocols for each participant.

Finally, the study tried to associate a very specific rather possibly dangerous outcome with the predictor of time of cord clamping for effective generation of clear and valid knowledge for program and implementation, unlike most of the studies in the area accompanying excessive outcomes, that can put proper implementation of study protocols at stake.

## 8.2. Limitations of the Study

The study has the following limitations that can be considered by the researchers and they might help for future advanced studies in the area:

- Due to limitation of resource, formulating the sample size relies on our secondary objective of "effect of time of cord clamping on jaundice requiring phototherapy" and it may lead to smaller sample size and reduce the probability of acquiring much more newborns in each group.
- The existing protocol of newborn care and postpartum hospital stay of the our study area (TASH) was not more than 24 hours and that limited our outcome data collection time to 24 hours only and that can minimize our probability of capturing the late (48-72 hours) complications related to bilirubin level like jaundice requiring phototherapy and can be considered for future studies in this area.

# 9. CONCLUSION AND RECOMMENDATIONS

## 9.1. Conclusion

This research finding found out and concluded that time of umbilical cord clamping will have no significant relationship with the total serum bilirubin levels of neonates at least within 24 hours of birth. In addition, delaying an umbilical cord clamping time to three minutes had no effect on elevating the total serum bilirubin levels of neonates that can put them in danger of jaundice requiring phototherapy with in the first 24 hours after birth.

## 9.2. Recommendations

The study findings indicate the following recommendations based on the evidences generated and categorized as follows:

**Health care providers**: based on these findings, it is recommended that newborn health care providers shall have no concern about exposing the neonates to unnecessary bilirubin related complications while providing a delayed umbilical cord clamping for better neonatal/infant nutritional and development outcomes, in addition, implementing the procedure is easily applicable that requires no additional resource except 2 – 3 more minutes of patience.

**Programmers/Policy makers:** In Ethiopia different programmers are trying to advocate the importance of delaying cord clamping time and they shall put much more efforts using these results and other multiple findings from studies as an advocacy tool to minimize the biases and perceptions among the health providers in Ethiopia.

**For professional Associations and Ministry of Health:** The concept of delaying umbilical cord clamping time is an old concept with new observations and it has a strong evidence-based recommendation in benefiting the neonates and/or infants. Furthermore, this study shall help the advocacy works on newborn health for positive outcomes.

**For researchers:** Neonatal health care researchers shall consider this study as an inception towards generating knowledge and clearing the myths that exist with the implementation of delayed cord clamping in our country Ethiopia and shall conduct a more structured randomized controlled trials towards similar other benefits and possible complications.

# 10.REFERENCES


1. UNICEF. Press-releases on global child birth report January 1, 2018 [cited 2018 10 September ]. Global child birth report. Available from: https://www.unicef.org/media/media_102362.html.
2. EPHI, FMOH. Ethiopian Emergency Obstetric and Newborn Care (EmONC) Assessment. Ethiopian public health institute; 2016 September 2017. Contract No.: 1.
3. WHO. Guideline: Delayed umbilical cord clamping for improved maternal and infant health and nutrition outcomes: World Health Organization; 2014.
4. Eichenbaum-Pikser G, Zasloff J. Delayed clamping of the umbilical cord: a review with implications for practice. Journal of midwifery & women's health. 2009;54(4):321-6.
5. Drummond G, Kappas A. Chemoprevention of severe neonatal hyperbilirubinemia. Seminars in perinatology. 2004;28(5):365-8.
6. Kamisako T, Kobayashi Y, Takeuchi K, Ishihara T, Higuchi K, Tanaka Y, et al. Recent advances in bilirubin metabolism research: the molecular mechanism of hepatocyte bilirubin transport and its clinical relevance. Journal of gastroenterology. 2000;35(9):659-64.
7. Watson R. Hyperbilirubinemia. Critical care nursing clinics of North America. 2009;21(1):97-120, vii.
8. Lazarus C, Avchen RN. Neonatal hyperbilirubinemia management: a model for change. Journal Of Perinatology. 2009;29:S58.
9. Vreman HJ, Wong RJ, Stevenson DK. Phototherapy: Current methods and future directions. Seminars in perinatology. 2004;28(5):326-33.
10. Greco C, Arnolda G, Boo N, Iskander I, Okolo A, Rohsiswatmo R, et al. Neonatal Jaundice in Low- and Middle-Income Countries: Lessons and Future Directions from the 2015 Don Ostrow Trieste Yellow Retreat. Neonatology. 2016;110(3):172-80.
11. Girma MTHHJP. Prevalence and Contributing Factors of Neonatal Jaundice in Neonatal Intensive Care Unit at St Paul's Hospital Millennium Medical College, Addis Ababa, Ethiopia, 2019. 2020;74.
12. Evans D. Neonatal jaundice. BMJ Clinical Evidence. 2007;2007:3-19.
13. Porter ML, Dennis BL. Hyperbilirubinemia in the term newborn. American family physician. 2002;65(4):599-606.
14. Greco C, Arnolda G, Boo NY, Iskander IF, Okolo AA, Rohsiswatmo R, et al. Neonatal Jaundice in Low- and Middle-Income Countries: Lessons and Future Directions from the 2015 Don Ostrow Trieste Yellow Retreat. Neonatology. 2016;110(3):172-80.
15. Cernadas JMC. Timing of umbilical cord clamping of term infants. Arch Argent Pediatr. 2017;115(2):1-7.
16. Mercer JS, Erickson-Owens DA, Collins J, Barcelos MO, Parker AB, Padbury JF. Effects of delayed cord clamping on residual placental blood volume, hemoglobin and bilirubin levels in term infants: a randomized controlled trial. Journal of Perinatology. 2017;37(3):260-4.
17. Andersson O, Hellström-Westas L, Andersson D, Domellöf M. Effect of delayed versus early umbilical cord clamping on neonatal outcomes and iron status at 4 months: a randomised controlled trial. BMJ. 2011;343(1):12.
18. BMJ. Neonatal jaundice_Best practice. British Medical Journal; 2018 June 30.
19. Organization WH. Guideline: Delayed umbilical cord clamping for improved maternal and infant health and nutrition outcomes: World Health Organization; 2014.
20. Darwin E. Zoonemia or the laws of organic life. Greg Alethoup, Robert Shimmin, Edkins

K, editors. London: The Royal Philosophical Society; 2005 May 1, 2005. 116-9 p.
21. Dunn P. Dr Erasmus Darwin (1731–1802) of Lichfield and placental respiration. Arch Dis Child Fetal Neonatal. 2003;88:4.
22. WHO. Beyond Survival: Integrated Delivery Care Practices for Long-Term Maternal and Infant Nutrition, Health and Development. Pan American Health Organization Washington, DC; 2013.
23. Farrar D, Airey R, Law G, Tuffnell D, Cattle B, Duley L, et al. Measuring placental transfusion for term births: weighing babies with cord intact. British Journal of Obstetrics and Gynecology. 2011;118(1):70-5.
24. James D, Carol H, Barry S. The quantitative assessment of body iron. blood. 2003;101:3359-64.
25. Saba K, Majeed T, Bukhari MH. Early versus Delayed Umbilical Cord Clamping Leads to Neonatal Anemia. ANNALS. 2012;18(3):309-15.
26. Ashish K, Rana N, Målqvist M, Linda J, Subedi K, Andersson O. Effects of Delayed Umbilical Cord Clamping vs Early Clamping on Anemia in Infants at 8 and 12 Months. JAMA Pediatrics. 2017;17(3):264-70.
27. Camila M, Lynnette M, Gilberto T, Raúl E, Kathryn G. Effect of timing of umbilical cord clamping on iron status in Mexican infants: a randomised controlled trial. The lancet. 2006;367:8.
28. Kelly Kuo, Priyanka Gokhale, David N, Chayatat Ruangkit, Bhola M, March M. Maternal outcomes following the initiation of an institutional delayed cord clamping protocol: an observational case–control study. The Journal of Maternal-Fetal & Neonatal Medicine. 2017:1-5.
29. Vasconcelos FB, Katz L, Coutinho I, Lins VL, AmorimMM d. Placental cord drainage in the third stage of labor: Randomized clinical trial. plos one. 2018;13(5):1-10.
30. Maria AM, Porter F, Chao TT-M. Delayed Umbilical Cord Clamping After Birth. Journal of Pediatrics. 2017;129(1):5-11.
31. Jane Houston, Lilla Dillon, Ashley Duvall, McGuire M. Bridging the Gap between Evidence and Practice: A Systematic Review—When Is the Best Time to Clamp the Infant's Umbilical Cord in Term Low-Risk Women? Open Journal of Nursing. 2014;4:730-36.
32. Saigal S, O'Neill A, Surainder Y, Chua L-B, Usher R. placental transfusion and hyperbilirubinemia in the premature. pediatrics. 1972;49(3):406-19.
33. Anna Locatelli, Consonni S, Ghidini A. Approach to Decreasing Complications of Prematurity. Obstet Gynecol Clin N Am. 2015;1(1):1-20.
34. Duley L, Batey N. Optimal timing of umbilical cord clamping for term and preterm babies. Early human development. 2013;89:905-8.
35. McDonald S, Middleton P. Effect of timing of umbilical cord clamping of term infants on maternal and neonatal outcomes. cochrane library. 2008;2008(2):1-29.
36. Pandey N, Gupta S, Yadav RK, Sarvottam K. Physiological Jaundice: Role In Oxidative Stress. Researchgate. 2013;05(19):69-80.
37. McDonald SJ, Middleton P, Dowswell T, Morris PS. Effect of timing of umbilical cord clamping of term infants on maternal and neonatal outcomes. Evidence-based child health : a Cochrane review journal. 2014;9(2):303-97.
38. Fogarty M, Osborn DA, Askie L, Seidler AL, Hunter K, Lui K, et al. Delayed vs early umbilical cord clamping for preterm infants: a systematic review and meta-analysis. American journal of obstetrics and gynecology. 2018;218(1):1-18.
39. Yao AC, Moinian M, Lind J. Distribution Of Blood Between Infant And Placenta After Birth. The lancet. 1969:871-3.
40. Olusanya B, Osibanjo F, Slusher T. Risk Factors for Severe Neonatal Hyperbilirubinemia in

Low and Middle- Income Countries: A Systematic Review and Meta-Analysis. plos one. 2015;10(2):1-16.
41. Stoll BJ, Kliegman RM. Jaundice and hyperbilirubinemia in the newborn. Nelson textbook of pediatrics 17th ed Philadelphia: WB Saunders. 2004:562-96.
42. Carvalho OMC, Augusto MCC, Medeiros MQ, Lima HMP, Viana Junior AB, Araujo Júnior E, et al. Late umbilical cord clamping does not increase rates of jaundice and the need for phototherapy in pregnancies at normal risk. The Journal of Maternal-Fetal & Neonatal Medicine. 2018:1-6.
43. Dennery PA, Seidman DS, Stevenson DK. Neonatal hyperbilirubinemia. New England Journal of Medicine. 2001;344(8):581-90.
44. Organization WH. Survive and thrive: transforming care for every small and sick newborn. 2019.
45. Ethipia MoH. National Strategy for Newborn and Child Survival in Ethiopia 2015/16-2019/20. In: Directorate MaCH, editor. 2016. p. 30 - 5.
46. Burke BL, Robbins JM, Mac Bird T, Hobbs CA, Nesmith C, Tilford JMJP. Trends in hospitalizations for neonatal jaundice and kernicterus in the United States, 1988–2005. 2009;123(2):524-32.
47. Lancet YICSSGJT. Clinical signs that predict severe illness in children under age 2 months: a multicentre study. 2008;371(9607):135-42.
48. ACOG, AAP. Delayed Umbilical Cord Clamping After Birth. Journal of Pediatrics. 2017:e20170957.
49. Cernadas JMC, Carroli G, Pellegrini L, Otan˜o L, Ferreira M, Ricci C, et al. The Effect of Timing of Cord Clamping on Neonatal Venous Hematocrit Values and Clinical Outcome at Term: A Randomized, Controlled Trial. pediatrics. 2006;117(4):779-86.
50. Rincon D, Foguet A, Rojas M, Segarra E, Sacristan E, Teixidor R, et al. [Time of cord clamping and neonatal complications, a prospective study]. Anales de pediatria (Barcelona, Spain : 2003). 2014;81(3):142-8.
51. Bhutani VK. Bilirubin nomogram, a prediction tool or natural history profile? Indian pediatrics. 2013;50(4):365-6.
52. Yu ZB, Han SP, Chen C. Bilirubin nomograms for identification of neonatal hyperbilirubinemia in healthy term and late-preterm infants: a systematic review and meta-analysis. World journal of pediatrics : WJP. 2014;10(3):211-8.
53. Ibrahim NO, Sukkarieh HH, Bustami RT, Alshammari EA, Alasmari LY, Al-Kadri HMJASM. Current umbilical cord clamping practices and attitudes of obstetricians and midwives toward delayed cord clamping in Saudi Arabia. 2017;37(3):216-24.
54. Leslie MS. Perspectives on ImplementingDelayed Cord Clamping. Jounnals CNE 2015;1(1):13.
55. Rana N, Brunell O, Målqvist MJPo. Implementing delayed umbilical cord clamping in Nepal—Delivery care staff's perceptions and attitudes towards changes in practice. 2019;14(6):e0218031.

# 11.ANNEXES

## Annex 1:Data Collection Tools (English)

### 1.1. Study informant sheet and consent form

**Introduction and objective**: Hello My name is _____. You are invited to participate in a research study on Time of cord clamping of newborns. From the information collected and studied in this project we hope to learn more about the most beneficial time of cord clamping among newborn babies, including possible related benefits and outcomes that will generate from it and you and your newborn are also selected to take part for that exact same reason.

**Procedures:** With your permission, we would like to collect health information about you and your newborn, including information about your general health (height, weight, blood pressure, results from blood tests, physical exam results) related current and prior medical history. Important information including Birthweight, Gestational age, sex, Apgar score at 1$^{st}$ minute of the newborn will be recorded from the chart, Physical examination or by asking you.

Additionally the study will have three groups with different observations that; Group 1 receive an immediate cord clamping(<30 seconds), Group 2 receive clamping at exactly 1 minute and the third group will receive cord clamping at exactly on 3 minutes. Your child will be assigned to any of the groups randomly. Before, during and after the cord management the newborn child will receive the Essential newborn care with the standard while the baby rests on your abdomen apart from the minute of cord clamping. After cord clamping you will continue to feed the newborn as usual and the midwife will monitor closely as the protocol of the hospital. Finally, the newborn will be examined visually for color of the skin before discharge to home or within 24 hours by the on call physician.

**Risk and benefits**

There will be no unmanageable risk and the only risk might be developing jaundice that can be treated with phototherapy. You might not receive any direct benefit from participation but the participation could highly benefit your current baby and future newborns of Ethiopia including your future babies if planned by preventing them exposed to iron deficiency anemia until the first 6 months and forth up to one year.

**Rights and confidentiality**

All of the procedures will be carried out by trained health professionals and you are allowed to stay at bedside until the data collection process is completed. Both your and your infant'smedical information that are going to be accessed forthis study remained confidential. The necessary data will be documented only using unique codes for each participant and no names or card numbers will be recorded. You have full right of accepting or denying of participating in this study without being forced by someone. You are free to cease the procedure at any time if you feel discomfort or due to any other reasons. Failure to participate or withdrawing from the study will not prevent your child from receiving the best medical care that the hospital provides. If you have any question regarding the information I gave you, please be free to ask me.

For further clarifications you can contact the coordinator of the research using the following phone number.

Research coordinator:Biruk Hailu …… Tel (0912128858)
Advisors: Dr. Mulugetabetre
Mr. Abiyseifu

Based on the information provided about the study, Are you willing to participate in the study?
**YES** **NO**
If the answer is **YES** please continue to the informed consent form below.

**Informed Consent Form**

I agreed to voluntarily participate in this study after fully understand the procedures that will be conducted.

Mother's/Companion's Signature_____

Date_____

## 1.2. Participant Identification

| | |
|---|---|
| Medical record number of neonate | _____/___ |
| Identification code of the neonate | _____ |
| Code of group | _____ |
| **Name of data collector** | |
| **Date of data collection (DD/MM/YYYY)** | ____/____/_____ |
| **Name of supervisor** | |
| **Date of supervision** | ____/____/_____ |
| **Group code**<br>**01 – ICC (<30 seconds) group**<br>**02 – DCC (30 -60 seconds) group**<br>**03 – DCC (≥180 seconds) group** | |

## 1.3. Maternal questionnaire

| **1.3.1. Maternal factors** | | | |
|---|---|---|---|
| Serial no | Questions | Response | SKIP |
| 101 | Age of the mother in years | ☐☐ | |
| 102 | Total number of pregnancy | ☐☐ | |
| 103 | Total number of Live births | ☐☐ | |
| 104 | Maternal blood group & RH factor | A- ……..01<br>A+ ……..02<br>B- ……03<br>B+ ……..04<br>O- ……..05<br>O+ ……...06<br>AB- ……07<br>AB+ ……08 | |
| 105 | Does she ever had any chronic Medical illnesses? | YES ……01 →<br>NO ……02 → | 106<br>107 |
| 106 | What type of medical illness? (Encircle one or more) | Hypertension……01<br>Diabetes mellitus…02<br>Hepatitis(A/B/C) …...03<br>HIV/ AIDS …….04<br>others (specify)_____05 | |
| 106 | What was the membrane status during admission? | Intact ……01<br>Ruptured…….02 | |

| | | | |
|---|---|---|---|
| 107 | Previous history of CS | 1.YES 2.NO | |
| 108 | Previous sibling history of Jaundice/hyperbilirubinemia | 1.YES<br>2. NO<br>3. unknown | |

## 1.4. Fetalrelated questionnaires

### 1.4.1. Fetal characteristics

| Serial no | Questions | Response | |
|---|---|---|---|
| 201 | Gestational age at birth (weeks) | ☐☐ | |
| 202 | Sex | Male……01<br>Female……..02 | |
| 203 | Birth weight (grams) | grams☐☐ | |
| 204 | 1st minute Apgar score | /10 ☐ | |
| 205 | Type of feeding preferred during hospital stay | Exclusive Breastfeeding……01<br>Exclusive Formula feeding…..02<br>Mixed feeding……..03 | |
| 206 | Neonatal feeding frequency (24 hours ) | <5x ……01<br>5—8x …….02<br>≥ 8x ……..03 | |
| 207 | Time of initiation of first feeding | ≤ 30 minutes……01<br>>30—60 minutes..02<br>>60 minute ……...03 | |
| 208 | Body temperature | Axillary temperature with in 15 minute…… | |

### 1.4.2. Medical factors

| | | | |
|---|---|---|---|
| 209 | Cord blood RBC count | ☐☐☐☐ | |
| 210 | Cord blood Cord blood Total serum bilirubin (mg/dl) | ☐☐ | |
| 211 | Newborn Blood group & Rh | A- ……..01<br>A+……..02<br>B-……03<br>B+……..04<br>O- ……..05<br>O+……...06<br>AB ……07<br>AB+ ……08 | |
| 212 | Any type of infection before or after delivery | TORCH……01<br>Chorioamnionitis…….02<br>Neonatal Sepsis……03<br>Others(specify)_____04 | |

| | | | |
|---|---|---|---|
| 213 | Any form of trauma during delivery | Bruising……01<br>Laceration…….02<br>Cephalo-hematoma.03<br>Others(specify)_____04 | |

## 1.5. Final outcome recording chart

| | | | |
|---|---|---|---|
| 301 | What is the mean total serum bilirubin level at the 24th hour after birth? | _____mg/dl | |
| 302 | Required phototherapy within 24 hours | YES …….01 →<br>NO……02 | 302 |
| 303 | Age in hours phototherapy started | hours ☐☐ | |
| 304 | Estimated blood loss during delivery | If <500mililiter(ml)……01<br>If 500-1000ml …… .02 }→<br>If ≥ 1000ml……03 | 304 |
| 305 | What was the cause of Postpartum bleeding? | Retained tissue……01<br>Trauma/injury …….02<br>Uterine atony……03<br>Thrombin(coagulopathy)….04<br>Others(specify) _____05 | |

**Annex 2:Amharic translated questionnaire**

**ክፍል 1 ፡የስምምነትመግለጫቅጽ**

***መግቢያእናየጥናቱአላማ፡***

ጤናይስጥልኝስሜ…….ይባላልከወሊድበኃላበሚሰራየጨቅላህጻናትእትብትእናየእንግዴልጅልየታእናጥቅሞቹዙሪያበሚሰራውበዚህጥናትእንዲሳተፉተጋብዘዋል፡፡
ከዚህከሚሰበሰበውመረጃእናጥናትለህጻናትበጣምጠቃሚየሚሆነውንየእትብትመለያየትደቂቃወይምጊዜለመለየትየምንጠቀም በትሲሆንአንቺናልጅሽምለዚህምርምርአስተዋጽዋቹከፍተኛእንደሚሆንእናምናለን.
**የመረጃአሰባሰብሂደት፡**በእርሶፍቃድበቅድሚያጠቃሚየሆኑእንደየእናትእድሜ፣የእርግዝናሁኔታ፣የህክምናታሪክ፣ የደምናየአካልምርመራውጤትየመሳሰሉትንእንዲሁምበህጻኑበኩልየወሊድክብደት፣የጽንሱእድሜ፣ጾታ፣የ1 ደቂቃአፕጋርውጤትእናየመሳሰሉትንየእርሶንፍቃድበመጠየቅየምንወስድይሆናል፡፡
ይህጥናትህጻናቱንበሶስትቡድንየሚከፍልሲሆን፣የእርሶምልጅከነዚህቡድኖችዉስጥበአንዱበእጣአማካኝነትየሚመደብሲሆን፣ **ቡድን 1** -ህጻኑበተወለደበ 30 ሰከንድዉስጥወይምወዲያዉኑእትብቱየሚለያይሲሆን፤**ቡድን 2** -ህጻኑበተወለደበ 1 ደቂቃ(60 ሰከንድ)ዉስጥየሚለያይይሆናል፤**ቡድን 3** - ህጻኑበተወለደበ 3ተኛውደቂቃ (180 ሰከንድ)ውስጥእትብቱይለያያል.ከወሊድበፊትበወሊድወቅትናከወሊድበኃላየእርሶህጻንአስፈላጊዉንየጨቅላህጻናትእንክብካቤየ ሚደረግለትሲሆንሁሉምአገልግሎትሲሰጥበእርሶእይታዉስጥየሚከናወንይሆናል፡፡
በተጨማሪምህጻኑአገልግሎቱንእየተቀበልናከዛምበኃላየተለማደውንየአመጋገብአማራጭመከተልየሚችሉይሆናል፡፡
በመጨረሻምእርሶናልጅዎወደቤትከመመለሶበፊትለልጅዎቀላልአካለዊምርመራበአይንብቻበመመልከትየሚደረግለትይሆናል፡፡
**ጥቅምናተጋላጭነትንበተመለከተ፡**
ይህጥናትየከፋናመቆጣጠርየማይቻልተጋላጭነትየሌለውሲሆንምናልባትምበልጅዎላይየቆዳቀለምቢጫመሆንየሚከሰትከሆነና ሌላምያልተገመተችግርከተፈጠረአስፈላጊዉንየህክምናእርዳታየሚያገኙይሆናል፡፡
ይህጥናትምናልባትምቀጥተኛጥቅምለእርሶባያስገኝምለሚወለደውልጅዎእንዲሁምለሌሎችየአገራችንወደፊትለሚወለዱህጻናት በመጀመሪያዎቹ12ወራትውስጥሊከሰትከሚችልየደምማነስበሽታየመከላከልጥልቅአላማአለው፡፡
በተጨማሪምለልጅዎየአእምሮእድገትእጅግአስፈላጊየሆነዉንየብረትንጥረነገርበበቂሁኔታእንዲያገኙየሚረዳይሆናል፡፡
***መብትናሚስጢራዊነትንበተመለከተ*** ፡
የመረጃአሰባሰብሂደትናጥናቱሙሉበሙሉበሰለጠኑባልሙያዎችየሚሰራሲሆንበመረጃአሰባሰብምወቅትበማርፊያዎት(አልጋዎት )ላይእረፍትእያደረጉመሳተፍናመከታተልይችላሉ፤
ከእርሶናከልጅየሚሰበሰበውመረጃሚስጢራዊነቱየተጠበቀናእያንዳንዱመረጃምበሚስጢራዊቁጥሮችብቻየሚመዘገብይሆናል፡፡
በዚህጥናትላይበራስዎትዉሳኔብቻየመሳተፍ፣
በፈለጉትጊዜየማቋረጥመብትእንዳሎትእንዲረዱናበማቋረጥዎምክንያትበእርሶምሆነበልጅላይየሚፈጸምምንምአይነትየአገልግሎ ትልዩነትእንደማይኖርእንዲረዱእንፈልጋለን፡፡

በመጨረሻምስለጥናቱምሆነሌላተያያዥጉዳዮችዙሪያጥያቄምሆነማብራሪያሲኖርዎትበቅርበትካገኙትየጥናቱአባልጋርመመካከር የሚችሉይሆናል፡፡

የጥናቱአስተባባሪስም፡ብሩክሃይሉተስፋዬ

ስልክ፡ 0912128858

- ❖ በቀረበሎትማስረጃመሰረትበዚህጥናትለመሳተፍፍቃደኛነዎት?

  **ሀ. አዎነኝለ. አይደለሁም**

- ❖ **አዎከሆነመልሶ፡- ከታችየሚገኘዉንስምምነትቅጽይሙሉ፡፡**

***<u>የስምምነትቅጽ</u>***

**በቂመረጃካገኘውበኃላበዚህጥናትላይለመሳተፍመስማማቴንበፊርማዬአረጋግጣለሁ፡፡**

**የእናትፊርማ ----------**

**ቀን ----------**

**ክፍል 2፡የተሳታፊመለያ**

| | |
|---|---|
| የህክምናካርድቁጥር | _____/___ |
| የጥናቱተሳታፊመለያቁጥር | _____ |
| የቡድንመለያቁጥር | _____ |
| የመረጃሰብሳቢስም | |
| መረጃውየተሰበሰበበትቀን (ቀን/ወር/ዓ.ም) | _____/_____/_____ |
| የተቆጣጣሪውስም | |
| ቁጥጥርየተደረገበትቀን | _____/_____/_____ |
| የቡድንመለያቁጥር<br>01 – ከግማሽደቂቃበታችየእትብትልየታ<br>02 – 1 ደቂቃየዘገየየእትብትልየታ<br>03 – 3 ደቂቃየዘገየየእትብትልየታ | |

| **2.1. የእናትመሰረታዊመረጃዎች** | | | |
|---|---|---|---|
| ተ.ቁ | ጥያቄ | መልስ | SKIP |
| 101 | የእናትእድሜበዓመት | ☐☐ | |
| 102 | ጠቅላላእርግዝናብዛት | ☐☐ | |
| 103 | ጠቅላላበህይወትየተወለዱህጻናትብዛት | ☐☐ | |
| 104 | የእናትየደምአይነትእናአርኤች | A- ……..01<br>A+ ……..02<br>B- ……03<br>B+ ……..04<br>O- ……..05<br>O+ ……...06<br>AB- ……07<br>AB+ ……08 | |
| 105 | ከዚህቀደምእናትማንኛዉምአይነትጽኑህመምታማጪቃልች? | አዎ……01<br>አላዉቅም ……02 | አዎከሆነመልሱ ወደጥያቄ 106 ይሂዱ |
| 106 | ምንአይነትጽኑበሽታ?<br>(ከአንድበላይማክበብይችላሉ) | የደምግፊት……01<br>የስኳርበሽታ…02<br>የጉበትቫይረስ(A/B/C)…..03<br>ኤችአይቪ/ኤድስ …….04<br>ሌላካለይጥቀሱ…… | |

| | | | |
|---|---|---|---|
| | | .05 | |
| 106 | ወደጤናተቋሙስትመጣየእንሽርትዉሃሁኔታምንነበር? | አልፈሰሰምነበር……01<br>ፈሶነበር…….02 | |
| 107 | ከዚህቀደምበአፕራሲዮንወልደሽታዉቂነበር? | 1.አውቃለሁ2.አላውቅም | |
| 108 | ከዚህቀደምከቤተሰብመሃልታሞሰዉነቱቢጫሆኖሚያዉቅ ሰውአለ? | 1.ነበረ<br>2.ኖሮአያዉቅም<br>3. አላስታዉስም | |

**2.2. የልጅመሰረታዊመረጃዎች**

| **ተቁ** | ጥያቄ | መልስ | |
|---|---|---|---|
| 201 | የእርግዝናእድሜ<br>(በሳምንታት) | ☐☐ | |
| 202 | የህጻንጾታ | ወንድ……01<br>ሴት……..02 | |
| 203 | የህጻንክብደት (በግራም) | ግ☐☐☐☐ | |
| 204 | የ1ደቂቃአፕጋርውጤት | /10 ☐ | |
| 205 | የአመጋገብምርጫበሆስፒታልቆይታወቅት | የእናትጡትወተትብቻ……01<br>የተቀመረ/ዱቄትወተትብቻ…02<br>የተቀላቀለአመጋገብ.……03 | |
| 206 | የልጅአመጋገብድግግሞሽ<br>(በ 24 ሰዓትዉስጥ) | ከ 5 ጊዜበታች ……01<br>ከ5 እስከ8ጊዜ……..……02<br>8 እናከዛጊዜበላይ……..03 | |
| 207 | ህጻኑከተወለደበኃላየመጀመሪያምግብየወሰደበትሰዓት | በ30 ደቂቃዉስጥ……01<br>ከ30 እስከ60 ደቂቃዉስጥ..02<br>ከ 60 ደቂቃበኃላ ……...03 | |
| 208 | የሰዉነትየሙቀትመጠን<br>（የብብትስርየሙቀትመጠንበ15 ደቂቃዉስጥየተለካ） | ☐☐ግሬድ | |

**2.3. ከህክምናጋርየተያያዙመረጃዎች**

| | | | |
|---|---|---|---|
| 209 | ከእትብትየተወሰደየቀይደምሴልመጠን | ☐☐☐☐☐ | |
| 210 | ከእትብትየተወሰደየቢሊሩቢንመጠን<br>(ሚ.ግ/ዴ.ሊ) | ☐☐ | |
| 211 | የልጅየደምአይነትናአር.ኤች | A- ……..01<br>A+……..02<br>B-……03<br>B+……..04<br>O- ……..05<br>O+……...06<br>AB ……07<br>AB+ ……08 | |
| 212 | ከወሊድበፊትወይምበኃላየተከሰተኢንፌክሽን<br>(ከአንድበላይመምረጥይቻላል) | ቶክሶፕላስሞሲስ/ሌሎችቫይረሶች/ሩቤላ/ሳይቶመጋሎ/ሃርፕስ….01<br>ኮሪዮ-<br>አምኒዮናይቲስ……..02<br>የጨቅላህጻናትኢንፌክሽን……03 | |

| | | | |
|---|---|---|---|
| | | ሌላ(ጥቀስ)_____.....04 | |
| 213 | በወሊድወቅትያጋጠመማንኛዉምዓይነትጉዳት (ከ1 በላይመምረጥይቻላል) | የቆዳመቀጥቀጥ/መቅለም……..01<br>የቆዳመቀደድ/ጭረት……02<br>ሴፋሎ-ሄማቶማ……..……03<br>ሌላ(ጥቀስ)……..04 | |
| **ክፍል 03፡ማጠቃለያየምርመራውጤትከ 24 ሰዓትበኋላ** | | | |
| 301 | ጠቅላላሴረምቢሊሩቢንመጠን | ሚግ/ዴ□□ | |
| 302 | በ24 ሰዓትዉስጥየፎቶ-ቴራፒህክምናተሰጥተል | አዎ ……01<br>አልተሰጠም……02 | መልሱአዎከሆነወደቀጣይጥያቄይሂዱ |
| 303 | የፎቶ-ቴራፒህክምናሲጀመርየልጅእድሜስንትነበር (በሰዓት) | □□ | |
| 304 | በወሊድወቅትየፈሰሰየደምመጠንበሞያዊግምት | ከ500 ሚሊሊትርበታች……01<br>ከ500 እስከ 1000ሚሊሊትር…..…02<br>ከ1000 ሚ.ሊትርበላይ……03 | መልሱኮድ 02/03 ከሆነቀጥሎሚመጣዉ ንጥያቄይመልሱ. |
| 305 | የደምመፍሰሱዋነኛምክኒያትምንነበር (ከ 1በላይመምረጥይችላሉ) | በማህጸንየቀረቲሹ……01<br>የመራቢያአካልጉዳት……02<br>የማህጸንጡንቻመላላት……03<br>ኮአጉሎፓዚ……….04<br>ሌላ(ጥቀስ)……..05 | |

**Annex 3: Standard operating Procedure (SOP)**

## 2.1. The consent procedure

Before initiation of the data collection the participant shall be informed well and reach at a decision only after signing an informed consent and the following procedures shall be followed accordingly:

- Make sure the information to be disclosed to the eligible participant at hand with the consent form.
- Even though the time and place make the discussion harder, try to do the discussion in private or on a less disturbing environment as much as possible.
- Ascertain participant's level of literacy. If illiterate, s/he should have a literate witness present and reasons for this should be explained. The participant may bring their own witness but when none is available, the site team may provide an impartial witness, who may be a member of the clinic staff that has no involvement with the study or a friend or family member of the participant. However, the designated person responsible for taking informed consent should ensure that there is no coercion from any witness chosen.
- Assure the participant that all communication between you, the subject and a witness, if applicable, is in complete confidence and trust driven.
- Ask the participant the language in which they prefer to communicate and use it, with the assistance of a translator, in addition to the witness if necessary, to continue counseling. The translator should not be a family member or a friend.
- Explain further using the participant information sheet:
- It is essential that the study be explained in detail by carefully explaining all aspects of the information sheet. This can be done by either reading the information about the study to the participant or by allowing the participant to read aloud.
- The purpose of the study, procedures involved in participation, number of days required for participation, schedule of visits, confidentiality, risks and discomforts, benefits and known side-effects and the right to refuse and withdraw at any stage of the study will be explained at length.
- Answer any questions the participant may have about study participation to their satisfaction.

- If the subject agrees, the relevant consent form will be signed and dated by the subject. This should also be signed and dated by the study staffs who have given the information and obtained the consent. The time for consenting should be recorded by both the staff and the patient.
- In case of illiterate subjects, thumb print of the subject will be accepted.
- If the potential subject cannot make decision during first interaction with the study staff, s/he should be given some time to think, discuss with the near and dear ones and decide.
- If the subject returns to the study center and decides to participate at a later date, the informed consent procedure will need to be repeated.
- No study procedure shall be undertaken before informed consent is being obtained. Once consent is obtained, move to the next step of identification of the participant mother information's either from her chart or interview.

## 2.2. Screening and Enrolment procedure

### 2.2.1. General procedures for enrolment of a subject:

- Make sure that the participant agreed to participate, signed and dated the Informed consent form.
- Assign or make sure a unique study number/code is assigned to the subject.
- Take complete medical history according to the screening form.
- Carefully check for eligibility and confirm based on the inclusion and exclusion criteria.
- Fill all relevant laboratory request forms before addressing the participant.
- Eligible participants shall be enrolled as soon as they are confirmed eligible and they will have explanation about the next procedure of the study and make sure they made an informed decision last time.

### 2.2.2. General criteria for screening a subject

The selection of the study participants should be based on the following criteria:

**Inclusion criteria**

- A live newborn with gestational age of 37-42 weeks diagnosed with first day of last menstrual period (LMP) or ultrasound.
- A live newborn who has no gross complications related with the pregnancy and delivery (including mal-presentation, fetal distress, and congenital malformations).
- A live newborn with whom the mother agreed and give informed consent.
- A live newborn with whom body weight is above 2500gm and below 4000gm at birth.

**Exclusion criteria**

The following neonates will be excluded at the appropriate time of diagnosis of the criteria.

- A mother with rhesus negative blood group or any medical or obstetric complications (hepatitis, HIV/AIDS, hypertensive disorders of pregnancy, diabetes and severe anemia).
- A newborn with known congenital malformation during pregnancy or delivery.
- Confirmed multiple pregnancy and mal-presentation.
- Neonate that require immediate resuscitation.
- Newborns with body weight of above 4100gm.
- Newborns with their gestational age at birth is >42 weeks.

## 2.3. Data collection procedures

### 2.3.1. Phase one

Before the start of data collection, the following procedures should be followed:

- Greet the participant and build a rapport very well.
- Make sure the participant signed the consent after an adequate information has been given.
- Fill out the participant identification form.

- Fill out the maternal questionnaire from her medical record or by interviewing her directly on the missing aspects.
- The standard care during and delivery will continue until the next phase of data collection immediately after expulsion of the fetus depending on the assigned group.

### 2.3.2. Phase two

- For all the newborns born from the eligible participants, take 1mililiter (ml) of cord blood and store it in the code labeled taste tube and the assistant will hand over the sample to family member with a laboratory request form to be transported to the laboratory.
- The requested tests will be serum bilirubin level, hematocrit, hemoglobin and blood group with Rh.
- The birth attendant will manage the clamping of the cord as the randomized assignment received from the supervisor and should not reveal it to the participant.
- At exactly one minute, the Newborn APGAR score will be measured and recorded later after the delivery by the assistant birth attendant.
- The standard newborn care will continue to be provided as per the hospital protocol.

### 2.3.3. Phase three

- Record the required information's on the fetal related questionnaire (i.e. fetal characteristics) as per the questions and their measurements.
- As soon as the laboratory result arrives, record results on the fetal related questionnaire (i.e. Medical factors section) as per the questions and their measurement scales.
- Finally, after 24 hours of delivery request another laboratory test (i.e. serum bilirubin level) to the newborn and record the result on the final outcome recording section of the data collection form with other questions that will be assessed together.
- Before discharge of the newborn, make sure all the questionnaires and forms are recorded completely.

## 2.4. Role of the project assistants around implementation of the study

### 2.4.1. Data collector roles

- Identify eligible participants based on the pre-determined criteria.
- Build a rapport, discuss about the study and help the participant sign an informed consent after clear understanding of the project.
- During phase one of the data collection, fill out the questions on the maternal questionnaire as per the steps.
- During phase two, the data collector will draw 1ml of cord blood for laboratory analysis and handover to the birth assistant.
- Manage the cord as per the category of time of clamping received from the supervisors.
- Measure and record the newborn APGAR score and record on the form.
- If an unexpected incident happens around the time of birth discontinue the data collection and report on the incident form later on.

### 2.4.2. Supervisors role

- Receive and store the randomization codes of the participant from the principal investigator in a secured place and procedure.
- Help the data collector in identification of the eligible participants and signing an informed consent.
- Make sure the information on phase one of the data collection filled on the forms correctly.
- Hand over the sealed envelope that contains assignment group of the participant as soon as the birth attendant prepares to assist the women deliver her baby.
- Arrange a digital clock with second counter and inform the birth attendant that the clamping of the cord be can done as per the categorization (except for the immediate clamping).
- Make sure the fidelity of the whole data collection procedure based on the pre-determined checklist by the principal investigator.
- Collect and submit the completed papers to the principal investigator (PI).
- Communicate about the study for any concerned body that request immediate explanation.

- Function as a communication bridge between the data collectors and the PI including payment/financial related aspects.

## 2.5. Randomization and masking

The randomization of study participants follows a series of technical procedures at different members of the project.

**Principal investigator/data manager**

- Generate a list of sequential number (e.g. from 001) for the protocol specified sample size and randomize for assignment of treatment sequence using a Microsoft Excel random number generator.
- Generate the randomization list, seal in an envelope and store on a safe place within the labor and delivery ward by identifying the order of the blocks that includes the participant numbers.
- Always assign the sequence of participants with ordered blocks; make sure that after finishing each block of participants continue the immediately following block number.

| Number | Sequence of randomization | |
|---|---|---|
| **1** | IMD DIM IMD IMD DIM MDI IMD DIM | Block 1 |
| **2** | IMD MDI IMD DIM MDI MDI IMD MDI | Block 2 |
| **3** | IMD IMD DIM DIM MDI IMD MDI IMD | Block 3 |
| **4** | MDI DIM DIM MDI DIM DIM MDI IMD | Block 4 |
| **5** | MDI DIM IMD MDI IMD MDI DIM DIM | Block 5 |
| **6** | DIM DIMDIM MDI DIM DIM MDI | Block 6 |

The randomization technique starts with random assignment of participants in to blocks and the order of the sequential assignment for the three groups of participants will be done using random sequence generator from Excel 2016 program available on Office 2016 package software (Microsoft Corporation).

**Annex 4: Protocol fidelity controlling checklist**

| S.no | Activities | Yes | No |
|---|---|---|---|
| **Phase one data collection** | | | |
| 1 | Does the data collectors greet the participant and build a rapport very well? | | |
| 2 | Does the participant sign a consent after through discussion and without pressure? | | |
| 3 | Was the participant informed with her level of literacy and language preference? | | |
| 4 | Does the collectors carefully check participants' eligibility? | | |
| 5 | Does all the inclusion and exclusion criteria reviewed before enrolling participants? | | |
| 6 | Does the collector fill out the maternal questionnaire properly? | | |
| **Phase two data collection** | | | |
| 1 | Does the randomized envelope with code readily availed? | | |
| 2 | Is the timer (Clock) ready? | | |
| 3 | Collect 1mililiter (ml) of cord blood and store it in a code labeled test tube (EDTA). | | |
| 4 | Does the above sample transport to laboratory with request paper within 30 minutes? | | |
| 5 | Does APGAR record at $1^{st}$ and $5^{th}$ minutes? | | |
| **Phase three data collection** | | | |
| 1 | Does the fetal related questionnaire record as per the questions and their measurements? | | |
| 2 | Does the laboratory result fetch and recorded timely before discharge of the newborn? | | |
| 3 | Collect 1mililiter (ml) of venous blood and store it in a code labeled test tube (EDTA). | | |
| 4 | Does the result transfer and received from the laboratory timely? | | |
| 5 | Does the maternal and fetal data collection forms fill out completely before discharge? | | |

**Annex 5: Bhutani Nomogram for risk assessment of neonatal serum bilirubin level**