

Proportion of Birth Asphyxia and Its Associated Factors among Newborns Delivered in Public Hospitals in Addis Ababa, Ethiopia, 2019: A Multicenter Cross-Sectional Study

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ABSTRACT

Background: Birth asphyxia is a serious clinical problem that causes neonatal mortality and morbidity worldwide, Even though Ethiopia has made significant progress in lowering the under-five mortality rate, the neonatal mortality burden has not decreased significantly, which may be due to birth asphyxia. Therefore, the purpose of this study was to address this gap by assessing the prevalence of neonatal asphyxia and its associated factors among newborns in Addis Ababa public hospitals, Ethiopia.

Subjects and Method: A facility-based cross-sectional study was conducted on 683 live newborns born at Addis Ababa public hospitals. Data were gathered through structured face-to-face interviews, questionnaire administration, and a systematic random sampling technique with proportional allocation. For statistical analysis, data were entered into Epi-data version 4.6 and exported to SPSS version 26. All variables with p-value ≤ 0.2 were taken into the multivariable model. A statistical significant level was declared at 95% CI and adjusted odds ratio (AOR) at a $p < 0.05$.

Results: The prevalence of birth asphyxia was 72 (10.6%) with a 95 % CI of (8.3 to 13.0). According to this study, birth asphyxia was significantly associated with the odds of newborns whose mothers were not educated (AOR= 8.09; 95 % CI= 1.63 to 40.19; $p= 0.011$), prolonged labor (AOR= 3.52; 95 % CI= 1.47 to 8.43; $p= 0.005$), ante-partum hemorrhage (AOR= 5.36, 95 % CI= 1.69 to 16.99; $p= 0.004$), preeclampsia (AOR= 5.09, 95 % CI= 2.09 to 12.47; $p < 0.001$).

Conclusion: Birth asphyxia was found in 10.6 % of neonates. Birth asphyxia was significantly predicted by the mother's educational level, complicated labor, and low birth weight.

Keywords: proportion, Birth asphyxia, Newborns, Addis Ababa.

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BACKGROUND

Birth asphyxia is defined by the World Health Organization (WHO) as the failure to initiate and sustain breathing at birth (Federal Minister of Health (FMOH), 2014). Birth asphyxia is defined as impairment in placental or pulmonary gas exchange that results in hypercarbia and hypoxemia. Birth asphyxia is the inability of newborns to cry or breathe normally after birth (WHO, 2012).

APGAR score of <3 for longer than 5 min; neurological manifestations and multi-system organ dysfunction (Haider and Bhutta, 2006). APGAR score may be depressed by not only by hypoxic-ischemic insult, but also by other non-asphyxia factors such as maternal analgesia, prematurity and infection. A prolonged depression of the APGAR score has been related with death or severe neurodevelopment outcome (Roberto et al., 2014). Any score <7 at 1, 5 and/ or at 10 min) is defined as Low APGAR scores or birth asphyxia (Mukhtar-Yola et al., 2018). Birth asphyxia is one of the top three causes of newborn deaths, along with infections and preterm birth complications, which account for 88 percent of newborn deaths in Africa, with birth asphyxia accounting for 24% (Haider and Bhutta, 2006). Studies show in Zambia, newborns with clinical birth asphyxia were 4.4 times more likely to have an abnormal neurologic examination than non-asphyxiated infants (Halloran et al., 2013). Babies born in sub-Saharan Africa have a very high risk of birth asphyxia because 280,000 deaths occur due to birth asphyxia during first day of life (Gebregziabher et al., 2018).

Birth asphyxia can occur suddenly, but there are often risk factors, such as antenatal factors such as hypertensive disorders, infections, or bleeding; fetal causes such as fetal growth restriction, breech presentation, or chronic fetal distress; or intra-partum factors

such as infections, placenta bleeding, uterine rupture, umbilical cord accidents, or prolonged labor (Kumar and Paterson-Brown, 2015; Wosenu et al., 2018). Birth asphyxia accounts 23% (29/1000) live birth neonates mortality in Ethiopia (Haider and Bhutta, 2006). Major causes of neonatal death are birth are asphyxia (23%), preterm complications (27%) and infections (36%) (Wayessa et al., 2012; FMOH, 2014). Therefore, this study is needed the risk factors for neonatal deaths are preventable and effective intervention that can prevent many of the deaths and reduce disabilities in survivors from birth asphyxia. Though Ethiopia has made significant progress in reducing under-five mortality, the neonatal mortality burden has not decreased as much, which may be attributed to birth asphyxia. In Addis Ababa there is a limited study found to assess issues on prevalence and associated factors of birth asphyxia. It will also help to improve the knowledge of health care providers and women about neonatal birth asphyxia during labor. So, the aim of this study is to address this gap by assessing the prevalence of neonatal asphyxia and its associated factors among newborns in Addis Ababa.

SUBJECTS AND METHOD

1. Study design

The study was conducted in Addis Ababa, which is the capital city of Ethiopia and Seat of African Union and the United Nations World Economic Commission for Africa. It covers an area of 527 square kilometers and has 10 sub cities. According to population projection value for 2019 the city has an estimated population of 4.592 million (World Population projections, 2019).

A Facility based cross-sectional study was conducted in Addis Ababa public Hospitals from November to December, 2019. The study was conducted in three Addis

Ababa public hospitals selected by lottery method. These selected hospitals were Gandhi Memorial hospital (GMH), Yekatit 12 hospital medical college (Y12HMC) and Zewditu Memorial Hospital (ZMH).

2. Population and Sample

Source and study population during the data collection period, all newborns delivered in Addis Ababa health facilities and all newborns delivered in public hospitals were included.

3. Study Variables

The dependent variable was birth asphyxia. The Independent variables: (1) Socio demographic variables of the mother: maternal age, occupation, marital status, educational status, residence and income; (2) Ante-partum variables: pregnancy condition: history of diabetes, preeclampsia, eclampsia, ante-partum hemorrhage and anemia, ANC follow up, previous still birth, gravidity and parity; (3) Intra-partum variables: prolonged labor, prolonged ROM, mode of delivery, meconium stained, cephalo-pelvic disproportion, fetal presentation and general anesthesia received during c-section and; (4) Neonatal variables: gestational age, prematurity and birth weight.

4. Operational Definition of variables

Birth asphyxia neonates born in the hospitals studied and diagnosed with Asphyxia by attending health professionals using an APGAR score of less than 7 at the 5th minute, unable to breathe or cry after birth and newborn done resuscitation.

5. Study Instruments

Data were collected using a closed-ended structured face-to-face interviewer-administered questionnaire and tools adapted from various literatures, as well as a review of the mother's profile card. To ensure consistency, the questionnaire was first developed in English, and then translated into Amharic and then back into English by

language experts. Trained data collectors interviewed participants at the appropriate location. Each hospital had three BSc midwives who were supervised by one senior BSc midwife. The principal investigator provided all necessary training.

A two-day training on data collection procedures was provided for those data collectors and supervisors, including how to conduct interviews, administer questionnaires, obtain consent, maintain confidentiality, and respect the rights of participants. The questionnaire was pre-tested on 5% of the outside sample to assess its completeness, clarity, length, skip patterns, and completeness of questioners.

6. Data Analysis

Following data collection, the data were manually cleaned, coded, and entered into Epi-Data version 4.6 before being exported and analyzed using SPSS Software version 26. To examine the relationships between dependent and independent variables, bi-variable and multivariable logistic regressions were used. All variables with p -value ≤ 0.2 were taken into the multivariable model to control for all possible confounders. To assess multi-collinearity in the regression, model the variance inflation factors (VIF) was conducted to identify correlation between independent variables. Hosmer and Lemeshow goodness of fit test was done to test the fitness of the model and it was found good. The cut point to declare the presence of statistical association between dependent and independent variable were p -value < 0.05 or AOR, 95% CI.

7. Ethics Clearance

The study received ethical approval from the University of Gondar's institutional review board (IRB). After explaining the purpose and procedure of the study, each respondent provided informed written consent. The instrument contained no name or other identifying information. To obtain

permission to conduct the study a formal letter was submitted to GMH, Y12HMC and ZMH. The eligible study participants were enrolled in the study only after they give written informed consent and were not forced to participate.

chosen for this study, with 678 (99.3%) newborn/ mother pairs participating. Approximately 199 (29.4%) of the mothers were between the ages of 25 and 29, with a median age of 27.7 years (SD was 5.11). The majority of respondents 463 (68.3%) were secondary and above educated, while 205 (30.2%) were primary educated, and 643 (94.8%) mothers were married. Participants' average monthly income was 5,076.53 and (SD 3,392.90) (Table 1).

RESULTS

1. Socio-demographic characteristics of the participants

Six hundred eighty-three live births were

Table 1. Socio-demographic characteristics of mothers whose newborns in selected public hospitals from November to December, Addis Ababa, Ethiopia, 2019 (n= 678)

Variables	Frequency (n)	Percentage (%)
Age_ group of the mother		
15-19	20	2.9
20-24	190	28.0
25-29	199	29.4
30-34	186	27.4
≥35	83	12.2
Marital status_ group		
Married	643	94.8
Un married	35	5.2
Educational status		
No education	15	2.2
Primary education	200	39.5
Secondary plus	463	68.3
Religion_ group		
Christian	499	73.6
Muslim	179	26.4
Occupation status		
Gov't employee	209	30.8
Private employee	173	25.5
Merchant	64	9.4
Housewife	232	34.2
Residency		
Urban	665	98.1
Rural	13	1.9
Income quartile		
<2700	163	24.0
2701-4495	172	25.4
4496-6000	188	27.7
>6001	155	22.9

2. Obstetrics and newborn factors of the birth asphyxia

Among respondents, 671 (99.0 %) mothers

visited health facilities during their pregnancy, with 465 (68.6%) visiting four or more times. The majority of 494 (72.9 %) visited health facilities during their pregnancy, with 465 (68.6%) visiting four or more times. The majority of 494 (72.9 %) visited health facilities during their pregnancy, with 465 (68.6%) visiting four or more times. The majority of 494 (72.9 %) visited health facilities during their pregnancy, with 465 (68.6%) visiting four or more times.

were mothers with newborns between 37 and 42 weeks of gestation. Membrane rupture occurred in approximately 649 (95.7 %) of cases in less than 18 hours. In this study, 200 (29.5 %) of mothers had complications

before or during delivery, including 45 (6.6%) anemia, 56 (8.3%) antepartum hemorrhage (APH), 97 (14.4%) preeclampsia, and 39 (5.8 %) eclampsia (Table 2).

Table 2. Obstetrics and newborn factors of birth asphyxia among mothers with newborns in selected public hospitals from November to December, Addis Ababa, Ethiopia, 2019 (n=678)

Variables	Frequency (n)	Percentage (%)
Parity		
Prmi	429	63.3
Multi	249	36.7
Mode of delivery		
SVD	327	48.2
Instrumentals	63	9.3
C/S	288	42.5
Did the mother receive analgesics	288	42.5
Yes	390	57.5
No		
Duration of ROM		
<18hour	649	95.7
≥18hour	29	4.3
Prolonged labor		
Yes	60	8.8
No	618	91.2
Anemia		
Yes	42	6.2
No	636	93.8
Ante-partum hemorrhage		
Yes	56	8.3
No	622	91.7
Preeclampsia		
Yes	97	14.3
No	581	85.7
Eclampsia		
Yes	39	5.8
No	639	94.2
Meconium-stained liquor		
Yes	87	12.8
No	591	87.2
Fetal distress		
Yes	86	12.7
No	592	87.3
Chorioamnionitis		
Yes	35	5.2
No	643	94.8

BW group		
<2.5	211	31.1
≥2.5	467	68.9

There were 72 cases of birth asphyxia (10.6%). The majority (51%) were unable to breathe, while 21 (3.1%) were experiencing

other symptoms such as inability to feed, bluish discoloration, abnormal movement, and others (figure 1).

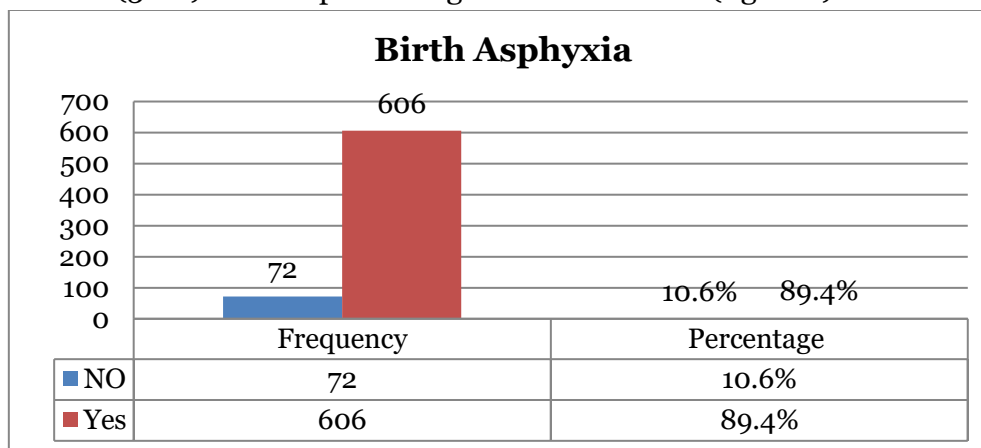


Figure 1. Distributions of birth asphyxia among newborns with mothers, at public hospitals, Addis Ababa, Ethiopia, 2019 (n= 678)

3. Associated Factors of birth asphyxia among newborns with mothers

In Multivariable analysis results showed that, there was statistically significance association found between birth asphyxia, parameters which showed p-value of below 0.05 were education of the mother, prolonged labor, APH, preeclampsia, fetal distress and Birth weight.

In this study, neonates whose mothers had a secondary education, the odds of birth asphyxia among newborns whose moms were uneducated were 8.09 (AOR= 8.09; 95% CI= 1.63 to 40.19; p= 0.011). Additionally, compared to neonates whose mothers had a secondary education level, those with a primary education level were 4.60 (AOR= 4.60; 95% CI= 2.07 to 10.23; p <0.001) times more likely to developed birth asphyxia.

In comparison to neonates whose mothers were born after complicated without ante-partum hemorrhage, those whose mothers were born after complicated with ante-

partum hemorrhage had a 5.36 (AOR= 5.36; 95% CI= 1.69 to 16.99; p= 0.004) times higher risk of developing birth asphyxia.

Contrarily, neonates whose mothers had preeclampsia complications were 5.09 times more likely to developed birth asphyxia than newborns whose moms did not have preeclampsia (AOR= 5.09; 95% CI= 2.09 to 12.47; p <0.001).

In this study, prolonged labor was a statistically significant contributor of newborn hypoxia. AOR= 3.52; 95% CI= 1.47 to 8.43; p= 0.005) times more likely to experience birth asphyxia in newborns born to mother who experience prolonged labor than their counterparts. Additionally, compared to neonates with a normal fetal heartbeat, those with intra-partum fetal distress were 3.99 (AOR= 3.99; 95% CI= 1.80 to 8.83; p= 0.001) times more likely to developed birth asphyxia.

The odds ratio of low birth weight was also a significant predictor of birth asphyxia.

Low birth weight babies were 3.86 (AOR= 3.86; 95% CI= 1.71 to 8.72; p= 0.001) times more likely to developed birth asphyxia than

babies with normal birth weight babies who weighed more than 2.5 kg (Table 3).

Table 3. Bivariate and multivariate logistic regression of birth asphyxia among newborns whose mothers at selected public hospitals from November to December, Addis Ababa, Ethiopia, 2019 (n=678)

Variables	Birth Asphyxia		COR (95% of CI)	P	AOR (95% OF CI)	P
	No (%)	Yes (%)				
Age group						
15-19	18 (90.0)	2 (10.0)	4.50 (0.59-34.11)	0.146	1.23 (0.09-16.02)	0.877
20-24	176 (92.6)	14 (7.4)	3.22 (0.72-14.51)	0.128	0.55 (0.10-2.96)	0.488
25-29	163 (81.9)	36 (18.1)	8.95 (2.10-38.08)*	0.003	0.90 (0.16-5.06)	0.905
30-34	168 (90.3)	18 (9.7)	4.34 (0.98-19.15)	0.052	0.63 (0.11-3.49)	0.596
≥35	81 (97.6)	2 (2.4)	1		1	
Education						
No education	11 (73.3)	4 (26.7)	4.59 (1.39-15.18)*	0.013	8.09 (1.63-40.19)**	0.011
Primary (1-8)	166 (83.0)	34 (17.0)	2.58 (1.56-4.29)*	<0.001	4.60 (2.07-10.23)**	<0.001
Secondary plus	429 (92.7)	34 (7.3)	1		1	
Income_ quartile						
<2700	136 (83.4)	27 (16.6)	5.96 (2.23-15.91)*	<0.001	0.88 (0.23-3.43)	0.852
2701-4495	142 (82.6)	30 (17.4)	6.34 (2.39-16.79)*	<0.001	2.24 (0.61-8.28)	0.226
4496-6000	178 (94.7)	10 (5.3)	1.68 (0.56-5.04)	0.350	0.66 (0.16-2.77)	0.566
>6001	150 (96.8)	5 (3.2)	1		1	
Parity						
Primiparous	391 (91.9)	38 (8.9)	0.62 (0.38-1.01)*	0.049	0.50 (0.24-1.04)	0.063
Multiparous	215 (86.3)	34 (13.7)	1		1	
Mode of delivery						
SVD	278 (85)	49 (15)	2.36 (1.37-4.08)*	0.002	10.09 (0.63-161.01)	0.102
Instrumentals	60 (95.2)	3 (4.8)	0.67 (0.19-2.33)	0.529	2.20 (0.13-36.34)	0.581
C/S	268 (93.1)	20 (6.9)	1		1	
Receive analgesics						
Yes	265 (92)	23 (8.0)	0.60 (0.36-0.98)*	0.058	8.01 (0.54-119.45)	0.131
No	341 (87.4)	49 (12.6)	1		1	
Rupture of Membrane						
<18 hour	585 (90.8)	64 (9.9)	0.29 (0.12-0.68)*	0.004	0.34 (0.09-1.22)	0.099
≥18 hour	21 (72.4)	8 (27.6)	1		1	
Prolonged labor						
Yes	31 (51.7)	29 (48.3)	12.51 (6.91-22.65)*	<0.001	3.52 (1.47-8.43)**	0.005
No	575 (93.0)	43 (7.0)	1		1	
Anemia						
Yes	32 (76.2)	10 (23.8)	2.89 (1.36-6.17)*	0.006	2.09 (0.53-8.38)	0.295
No	574 (90.3)	62 (9.7)	1		1	
APH						
Yes	40 (71.4%)	16 (28.6)	4.04 (2.13-7.68)*	<0.001	5.36 (1.69-16.99)**	0.004
No	566 (91.0)	56 (9.0)	1		1	
Preeclampsia						
Yes	64 (66.0)	33 (34.0)	7.17 (4.21-12.19)*	<0.001	5.09 (2.09-12.47)**	<0.001
No	542 (93.3)	39 (6.7)	1		1	
Eclampsia						
Yes	14 (48.3)	15 (51.7)	11.13 (5.11-24.21)*	<0.001	1.96 (0.59-6.49)	0.271
No	592 (91.2)	57 (8.8)	1		1	
Meconium stained liquor						
Yes	56 (64.4)	31 (35.6)	7.43 (4.32-12.76)*	<0.001	1.05 (0.42-2.64)	0.924
No	550 (93.1)	41 (6.9)	1		1	
Fetal distress						
Yes	58(67.4%)	28 (32.6)	6.01 (3.48-10.38)*	<0.001	3.99 (1.80-8.83)**	0.001
No	548(92.6%)	44 (7.4)	1		1	
Chorioamnionitis						
Yes	22 (62.9)	13 (37.1)	5.85 (2.80-12.21)*	<0.001	2.64 (0.82-8.51)	0.104
No	584 (90.8)	59 (9.2)	1		1	

Birth weight group

<2.5	161 (76.3)	50 (23.7)	6.28 (3.69-10.70)*	<0.001	3.86 (1.71-8.72)**	0.001
≥2.5	445 (95.3)	22 (4.7)	1		1	

N-Observation=678

-2 log likelihood=251.57

Nagelkerke R²= 53.6%

Keys: 1= Reference

*P<0.20, COR, statistically significant by bivariate analysis

**P<0.05, AOR, statistically significant by multivariate logistic regression

DISCUSSION

In this study, the prevalence of birth asphyxia among newborns delivered in public hospitals in Addis Ababa was 10.6% with 95% CI (8.3 to 13.0). This finding is lower compared to a study conducted in Debre Tabor hospital, Ethiopia (29.9%) (Asfere and Yesur, 2018), in Jimma hospital Southwest, Ethiopia (32.9%) in the first minute and (12.5%) in the fifth minute (Zelalem et al., 2018) and in Nigeria (21.1%) (Ajao and Adeoye, 2019). In addition, this finding is lower when compared to a systematic review which was conducted in Debre Markos University, Ethiopia (21.0%) (Sandeku et al., 2020). This variation may be due to the fact that this study included more than 98.8 % of newborns with mothers who visited antenatal care. Furthermore, in Addis Ababa, good infrastructure, easily accessible health facilities, and family awareness may have a positive influence on the lower prevalence of birth asphyxia in our study. However, it was higher than in a previous study conducted at Dil-chora referral hospital in Dire-dawa, Ethiopia (3.1%) (Ibrahim et al., 2017). This explains why institutional deliveries may be self-referred obstetric complications or delays in health centers with high neonatal complications.

One of the factors associated with birth asphyxia in this study was maternal education. When compared to secondary plus education levels, non-educated mothers were 8 times more likely to develop birth asphyxia. This finding is consistent with studies conducted in Tigray (Tasew et al.,

2018), Dire-dawa (Ibrahim et al., 2017), Kenya (Kibai, 2017), Tanzania (Schmieglow et al., 2014), Nepal (Anne et al., 2019) and Sweden (Milsom et al., 2012). This is justified by the fact that uneducated mothers are a broad indicator of poor socioeconomic conditions, which are associated with subsequent malnutrition, frequent pregnancies, and influence care seeking during the postpartum period. According to this study, being illiterate has a negative relationship with antenatal care follow-up, which has a negative impact on birth asphyxia.

Ante-partum hemorrhage was found to be significantly associated with birth asphyxia. Mothers who had ante-partum hemorrhage had a 5 times higher risk of birth asphyxia than those who did not have ante-partum hemorrhage. This study is in line with studies reported previously in Tigray (Tasew et al., 2018), Dire-dawa, Ethiopia (Ibrahim et al., 2017) and Pakistan (Kiyani and Khushdil, 2014). This could be because there is less blood flow from the mother to the placenta during antepartum bleeding, causing hypoxemia in the fetus. If the mother's transfusion or delivery is delayed, this condition can result in perinatal asphyxia.

When compared to non-asphyxiated newborns, newborns whose mothers had Preeclampsia were 5 times more likely to develop Birth Asphyxia. Furthermore, this finding is consistent with findings from a study conducted in Dire-dawa referral hospital in Eastern Ethiopia (Ibrahim et al., 2017), in Jimma zone public hospitals, Southwest Ethiopia (Wayessa et al., 2018),

in Nigeria Public Hospital, Nigeria (Ajao and Adeoye, 2019) and in Karachi Hospital in Pakistan (Kiyani and Khushdil, 2014). Preeclampsia have been associated with increased risk of birth asphyxia in multiple hospital-based studies in Nepal (Anne et al., 2019). Births that occur as a result of a complicated labor, such as Preeclampsia, may be affected in a variety of ways, increasing their risk and resulting in vulnerable babies who are prone to asphyxia when compared to uncomplicated labor. This is because labor complications, such as preeclampsia-related issues, can reduce the infant's blood and oxygen supply, leading to birth asphyxia.

Prolonged labor was found to be a statistically significant risk factor for birth asphyxia in this study. Birth Asphyxia was 4 times more likely in newborns delivered after a long labor. This finding agrees with findings of in Tigray (Gebregziabher et al., 2018), Gondar (Wosenu et al., 2018), Dire-dawa (Ibrahim et al., 2017) and Pakistan (Kiyani and Khushdil, 2014). This could be because prolonged labor causes the baby to be involved in labor for a longer period of time, which increases the risk of birth trauma; additionally, many conditions associated with prolonged labor make newborns more likely to be exposed to Pitocin and instrumental assisted deliveries. All of this can lead to the baby suffering from birth asphyxia. Another explanation is that if the labor does not proceed normally, the woman may suffer from serious complications such as dehydration, exhaustion, or uterine rupture. It is also clear that when labor is prolonged, the fetus is more likely to become distressed.

Intra-partum fetal distress was another neonatal factor found to be significantly associated with birth asphyxia. Neonates born with normal fetal heart rate had a lower risk of birth asphyxia than those born with

intra-partum fetal distress. A similar result was obtained in a previous study done in Gondar, Ethiopia (Wosenu et al., 2018). The possible explanation is that fetal distress is the main indication of emergency CS which is a known risk factor for birth asphyxia.

Low-birth weight was also a significant predictor of birth asphyxia in this study. Low birth weight neonates were 4 times more likely to suffer from birth asphyxia than normal birth weight neonates. This finding was in line up with those studies conducted In Jimma zone public hospitals (Zelalem Wayessa¹ et al., 2018), Tigray (Gebregziabher et al., 2018), Dilla (Alemu et al., 2019), University of Gondar Referral Hospital (Worku and Yusef, 2017) and Pakistan (Kiyani and Khushdil, 2014). This could be explained by the fact that a high proportion of low-birth-weight babies are pre-term, which causes them to lack surfactant, resulting in difficulty breathing and subsequent birth asphyxia.

One of the most common causes of neonatal morbidity and mortality in the labor and delivery ward is birth asphyxia. As a result, efforts should be made to improve the quality of intra-partum care service in order to prevent prolonged labor and fetal complications, as well as to identify and closely monitor mothers who are experiencing fetal distress. Because the prevalence of birth asphyxia was 10.6% in this study area, we recommend that proper emphasis be given to neonatal mortality by addressing and providing adequate information.

AUTHOR CONTRIBUTION

All authors should have made substantial contributions to all the following: the conception and design of the study, or acquisition of data, or analysis and interpretation of data, drafting article or revising it critically for important

intellectual content, final approval of the version to be submitted.

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This study used the researcher's personal funds.

CONFLICT OF INTERESTS

The authors declare no conflict of interest.

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