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Bacterial Neonatal Sepsis and Associated Risk Factors among Neonates: The Case of Adama Hospital Medical College

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Abstract

Background: Bacterial neonatal sepsis is a systemic inflammatory response to infection and/or isolation of bacteria from the bloodstream in the first 28 days of life.

Objective: the aim of this study is to assess neonatal sepsis and associated factors among neaonates admitted to Adama Hospital Medical College, Ethiopia

Methods: A cross-sectional study was conducted on 333 neonates, consecutive sampling used on neonates admitted at intensive care unit in Adama Hospital Medical College, Adama, Ethiopia. Data were collected using semi-structured questionnaire and 1ml of blood sample was collected following standard aseptic techniques and inoculated into a BACTEC Peds Plus culture vial, and transported to referral laboratory. Gram staining and sub-culturing were performed for each presumptive positive vial. Bacterial isolation and identification were done by enzymatic and fermentative biochemical tests and analyzed using SPSS v.20. multivariable logistic regression was used to identify factors having significant association with neonatal bacterial sepsis and 95% CI and P-value <0.05 were to state significance level of variable.

Results: Of the 317(95%) collected blood specimen, 147(46.4%)(95% Cl, 41.8 – 52.8%) showed bacterial growth. Gram-negative bacteria were dominants (69.9%); K. pneumoniae 72(48.98%), coagulase negative Staphylococci species 23(14.97%) and E. cloacae 18(12.24%). Factors like Onset of Labor(AOR=2.70, 95% CI: 2.34, 25.41), history of UTI/STI Infection (AOR=3.73,95%CI: 1.35, 10.28), Neonate age 4-7 days and 8-28 day (AOR=4.30, 95% CI: 1.91, 9.65 and AOR=6.07, 95%CI: 2.02, 18.22 respectively) low birth weight (AOR=2.24, 95% CI: 1.1.06, 4.71), Gestation age (AOR=2.53, 95% CI: 1.19, 5.38), intravenous fluids medication (AOR= 5.21, 95% CI: 2.58, 10.51), resuscitation at birth (AOR=7.85, 95% CI: 3.50, 17.58), shows significant association with neonatal sepsis.

Conclusion: The magnitude of bacterial sepsis was considerably high. Factors like onset of labor, history of UTI/STI infection , neonate age 4-7 days and 8-28 day, low birth weight, gestation age, intravenous fluids medication, resuscitation at birth, shows significant association with neonatal sepsis. Therefore, adherence to strict aseptic techniques while providing neonatal resuscitation and intraveinus fluid and medication service is important to reduce neonatal sepsis. [*Ethiop. J. Health Dev.* 2023; 37(1) 000-000]

Key words: Blood culture, Neonatal sepsis, Adama, Ethiopia

Introduction

Bacterial neonatal sepsis is a systemic inflammatory response to infection and/or isolation of bacteria from the bloodstream in the first 28 days of life and is a major cause of morbidity and mortality in newborns (1, 2). Neonatal sepsis is classified into two major categories based on the time of onset: early-onset sepsis (EOS) and late-onset sepsis (LOS) (3). EOS refers to sepsis in neonates at or before 72 hours of life, and LOS is defined as sepsis occurring after 72 hours of life. Typically, early-onset sepsis is considered maternally-acquired, usually from the maternal genital tract, and late-onset sepsis is generally regarded to originate from the care-giving environment, either a health-care or community setting (2,4).

Globally, WHO estimates that nearly 1 million neonatal deaths occur yearly, which accounts 10% of underfive mortality and 42% the neonatal death occurs in the first week of life of neonate in sub-saharan Africa (5). In these countries neonatal sepsis is responsible for 30-50% of neonatal deaths (6). In Ethiopian, about 37% neonates die from bacterial sepsis, which accounts for more than one third of neonatal deaths (7-9). Cross-sectional study conducted in Africa: Lusaka, Zambia, 33% (10), Dar'es Salaam, Tanzania 31.4%, Asmara, Eritrea 17% (11) and Khartoum, Sudan 67.5%(12) of neonates were diagnosed cultural confirmed bacterial sepsis. Neonatal sepsis reported in Ethiopia: 32.1% from Dilchora Hospital, Dire Dawa (13), 46.6% from Gondar Hospital (14) and 44.7% from Addis Ababa (15).

Studies from Asian region, reported gram-negative bacteria (>60%) as a major cause of neonatal sepsis. Study from Eastern India in the period from 2007-2009 reported gram-negative bacteria was predominant (71.6%), with K. pneumoniae being the most common isolate (16). Followed by Proteus spp.(26.3%), E. coli (21%), Pseudomonas spp. (18.4%), S. aureus (5.2%) and Salmonella spp.(2.6%) (17). Similarly, studies from four Arab countries, Klebsiella spp, Serratia spp.,

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Enterobacter spp., Escherichia coli, and Pseudomonas spp. being the most frequent bacteria isolates. Interestingly, gram positives like CoNS, and S. aureus and, Group B Streptococci were the predominant pathogen in Saudi Arabia, Bahrain and Kuwait and, in the United Arab Emirates, repectivley (18). In some of the Ethiopian studies, gram negative was the dominant isolates among septic neonates; Dire Dawa (13) and Addis Ababa (15) K. pneumoniae followed by S. aureus. In another studies gram positives were dominant bacterial isolates (14, 19).

Early newborn infections are frequently related to labor and childbirth and are caused by an entirely different spectrum of pathogens than the late neonatal infections (20). Several studies across the world reported that; maternal fever, Prolonged Rupture of Membrane (PRoM), and Urinary Tract Infection (UTI) or Sexually Transmitted Iinfection (STI) are the main maternal risk factors associated with neonatal sepsis (21). A study done in Cipto Mangunkusumo Hospital, Jakarta, Indonesia, showed that risk of neonatal sepsis was higher in PRoM for ≥ 18 hours before hospital admission, PRoM for ≥ 15 hours during hospitalization, and PRoM for ≥ 48 hours until birth (22). A neotate born from women with history of UTI/STI were 3.99 to 5.0 times (23-25) more likely to develop EONS (25).

The main neonatal risk factors for sepsis were, low birth weight and gestational age (preterm) (26). Neonates weight less than 2.5kg (23). Systematic review in India revealed that, gestational age of the newborn was significantly associated with the likelihood of neonatal sepsis, specifically newborn gestation <37 weeks (27), the type of bacteria is could also be the cause for neonatal sepsis (28). The baby care-related factors like mechanical ventilation, invasive procedures, and administration of parenteral fluids can predispose neonatal sepsis. On the other hand, poor hygiene, poor cord care, and bottle feeding, rather than breastfeeding are conditions that might fuel the risk of community-acquired neonatal sepsis (4).

In general, according to different studies reviewed above, the most common cause of EOS is vertical infection associated to maternal related conditions whereas, LOS is usually occurred via horizontal transmissions, as a result of hospital-acquired infection among high-risk neonates or community-acquired an infection that may be related to different neonatal care practice. Ethiopian government is working to reduce the prevalence of neonatal sepsis by encouraging institutional birth and creating awareness to women about sign and symptons of neonatal infection for each women during ANC service provision. As to the knowledge of researchers, there is study conducted to indicate the reason for neonatal sepsis in the study arera. Therefore, this study will help to explore factors related to neonatal sepsis

MATERIALS AND METHODS

Study area and period

Data was collected in Adama Hospital Medical College (AHMC) form October 1, 2020 to December 31, 2020, Adama town, Ethiopia. AHMC is one of the four hospitals based medical colleges in Ethiopia. The Hospital is serving a catchment population of more than 6 million; from four regions (Oromia, Amhara, Afar, and Somali). The Hospital has 298 beds capacity and serving on average 1000 patients/day at six medical case teams and different specialty clinics and six in-patient department. The NICU ward has 95 beds currently staffed with pediatricians, general practitioner, neonatal nurses and newborn care trained clinical nurses.

Study design

A facility based cross-sectional study design was implemented on neonates admitted in NICU at AHMC.

Source population

The source population includes all neonates admitted to AHMC.

Study population

The study populations includes all neonates admitted to neonat ward of AHMC during study period.

Inclusion and exclusion critetia

Inclusion criteria

All neonates admitted to AHMC neonatal ward and clinically diagnosed as sepsis was included into this study.

Exclusion criteria

All neonat admitted to AHMC neonatal ward and started antibiotic five days before the start of this study will be excluded from the study.

Neonates, who were not clinically diagnosed as sepsis and were on antibiotic for more than 5 days before date of data collection, were excluded from the study. Neonates were recruited based on clinical diagnosis to propabale sepsis and included in the study consecutively. A neonate on antibiotic treatment 5 days before blood sample collection were excluded.

Sample size determination

Sample size was determined by using single population proportion formula and the sample size for bacterial agents was calculated by taking prevalence of culture-proven neonatal sepsis in Ethiopia. The sample size of a study conducted at Dilchora Hospital (32.1%) (13) by considering, margin of error (d) 5% with the confidence interval level of 95%, and 10% non-response rate was 369 neonates. Since the study population, total number neonates admitted to hospital in one year was 3,418, which is <10,000. Finite population correction formula was used to determine the minimum sample size, which became 333 neonates.

Sampling procedure

Neaonates admitted to AHMC neonatal ward were included and sampled. Consecutive sampling procedure was used to select the study units. The sampling procedure continued until the required sample size was achieved. Newly admitted neonates were selected purposely to exclude selection of neonates who are on antibiotics for the last 5 days.

Data and blood sample collection

A semi-structured interviewer-administered pre-tested questionnaire was used to collect the data, adapted from literatures (13, 15). Data collection tool was

prepared in English and then translated to the local languages (Amharic and Afan Oromo) and then back to English for checking consistency of the questionnaires. Data was collected during the admission and by reviewing the registration book records in the labor ward, NICU, and gynecologic ward.

After cleaning the venous site with 70% alcohol and subsequently by 10% povidone-iodine solution, about 1 milliliter of blood was taken aseptically from a peripheral vein as described by Hall and Lyman (29) and injected into the BACTEC Peds Plus culture vials. The bottles was labeled with a unique sample number, date and time of collection, then immediately transported to Bacteriology Department of Adama Public Research and Referral Laboratory Center. The inoculated vials was incubated in an automated BACTEC system at 35 ± 2°C for 5 days as per manufacturer's instructions.

Bacteria isolation and identification

A positive result was indicated by an audible alarm and yellow illumination of the positive indicator lamp at the site of the positive vial in a BACTECTMFX40 system. On the computer, instrument status displays the station number was shown by flashing green in case of a positive vial. The bottles were incubated for 5 days before being reported as negative. Blood agar, chocolate agar, MacConkey agar culture media (from Oxoid, UK) were used to subculture from each presumptive positive vial (30). The blood and MacConkey agar plates were incubated aerobically while chocolate agar plates were incubated in a candle jar (at 5% CO_2 atmosphere) and examined for growth after 24–48 hours of incubation.

Bacterial isolates was identified by standard microbiological techniques (gram staining, colony characteristics, and biochemical testing). Gram-positive bacteria were confirmed by biochemical tests which include coagulase and catalase, growth on mannitol salt agar, and hemolytic activity on blood agar, bile aesculin test, bacitracin and optochin susceptibility test. For gram-negative bacteria, indole production, motility, citrate utilization, urease production, triple sugar iron, lysine iron agar and oxidase tests (from Deben Diagnostics, UK and Oxoid, Australia) were performed (31).

Variables

Dependent Variable: Neonatal sepsis is dependent variable

Independent variable:

Maternal related variables: Age, Educational status, Parity, ANC Visit, History of having STIs, Prior maternal infections, Onset of labour, Duration of labor, Duration of rapture of membrane, Amniotic fluid infection etc

Neonate related factors: Age, Sex, Gestational age, Multilple birth, Meconium stained, Birth weight, Breast feeding etc

Health care service related factors: Place of delivery, Mode of delivery, Resuscitation, Duration of admission, IV treatment procedure etc

Others: bacterial type

Quality Control and Assurance

Data was collected by two trained neonatal nurses. Then the checklist was pre-tested at similar Hospital on 5% of the total sample size 15 days before the data collection day. To ensure the accuracy of data, double data entry method was used (data was entered into epi info version 7 and then exported to SPSS version 20).

During laboratory analysis, pre-analytical, analytical and post-analytical standard operating procedures of the medical microbiology laboratory were followed. Culture media were prepared and sterilized based on the manufactures instruction. Sterility and performance of culture media was checked by incubating 3-5% prepared media for each batchs at 35-37 ^{0}C for 24 hours. The American Type Culture Collection (ATCC) reference strains which include E. coli (ATCC-25922), S. aureus (ATCC-25923), P. aeruginosa (ATCC-27853) and N. gonorrhoeae (ATCC-49226) for fastidious bacteria, all strains were obtained from Ethiopian Public Health Institute (EPHI) and used as quality control throughout the study for culture, biochemical and antimicrobial susceptibility testing.

Data processing and analysis

A standard coding guide, data entry, and detailed computer editing specifications were prepared and the data was entered into the computer using Epi info version 7 software and exported to SPSS 20.0 for cleaning and data analysis. Bivariate and multivariate logistic regression analysis were performed to evaluate whether individual predictors of interest were independently significantly associated with confirmed sepsis. Variation inflation factor was used to assess the multicollinearity and Hosmer and lemshow test was used to test normality of data. Only variables with significance $P < 0.25$ at a 95% confidence interval in the bivariate analysis were considered for inclusion in the multivariable analysis. Adjusted OR with its 95% CI were used as a measure of association between predictor variable and the outcome variable and $P < 0.05$ in the multivariable analysis were considered significant. Finally the data were presented in statement, table and graph.

Ethical issues

Ethical clearance was obtained from AHMC Ethical Review Board (Ref. No, 0382/K-373/12) and the Oromia Health Bureau support letter (Ref.No, BEFO/HBTFU/1-16/353) was communicated to AHMC and the center where the pre-test was performed. Informed consent was obtained from each parent/guard before the initiation of data collection.

Results

Socio-demographic characteristic of the study participants

This study enrolled 317 participants (95% response rate). The mean age of mothers were 26 years (SD±5) and nearly two third of them were 25-35 years old, and they were urban residents. Regarding maternal education, 145 (45.7%) attended secondary school and more than 42% did not attended beyond primary

school. Almost half 159 (50.2%) of mothers were primipara, about 221 (69.7%) had 1-3 antenatal care follow-up during their pregnancy, one third had labor of 6-12 hours and 277 (87.4%) gave birth with spontaneous vaginal delivery. Concerning the rapture of membrane, nearly three quarters of the mothers stayed 18 hours or less with membrane rapture before giving birth and 269 (84.9%) their amionitic fluid has no foul smell. About 53 (16.7%) of mothers had history UTI/STI during gestational period and 61 (19.3%) of mothers had fever. At the time of birth stained meconium was observed in 256 (80.7%) of neonate (Table 1).

Table I: Sociodemographic and reproductive related characteristics of mothers and neonatals admitted in NICU, AHMC, (n = 317)

Variable	Frequencies	Percent(%)
Mother age (years)		
15-24	100	31.6
25-35	209	65.9
>35	8	2.5
Residence		
Urban	212	66.9
Rural	105	33.1
Education Status		
Illiterate	40	12.6
Primary school	95	30.0
Secondary school	145	45.7
College and Higher	37	11.7
Parity		
Primipara	159	50.2
Multipara	158	49.8
ANC visits		
1-3 visits	221	69.7
≥4 visits	92	29.0
No visit	4	1.3
Onset of labor		
Spontaneous	277	87.4
Induced	40	12.6
Duration of labor (hrs)		
<6	111	35.0
6-12	95	30.0
13-24	77	24.3
>24	34	10.7
Membrane rupture duration (hrs)		
≥18	65	20.5
<18	232	73.2
Not ruptured	20	6.3
Amniotic fluid foul smelling		
Yes	48	15.1
No	269	84.9
Meconium stained		
Yes	67	21.1
No	250	78.9
History of UTI/STI		
Yes	53	16.7
No	264	83.3

Fever during labor		
Yes	61	19.2
No	256	80.8

Key: ANC-Antinatal care; UTI -Urinary tract infection; STI- Sexually transmitted infection

Neonate related factors

More than two third 213 (67.2%) admitted to hospital due to sepsis were found at the age of less than or equal to 3 days 213 (67.2%) with the median age of 1 day with the IQR of zero. Of the total neonates, 202 (63.7%) were male. About one third of neonate 97 (30.6%) were preterm and 25 (7.9%) were multiple birth. About 169 (53.3%) neonates were low birth weight, the majority of neonate 307 (96.8%) were born in health facilities and about two third 202 (63.7%) had spontaneous vaginal delivery.

Concerning the the clinical situation of neonate on birth, only one fifth 65 (20.5%) were resuscitated. The majoriy 303 (95.6%) of neonates blood sample was taken with in 48 hours of admission and 211 (66.6%) had exposure to invasive procedures before blood sample is taken. Almost half 167 (52.7%) of neonated initiated to breast feed with in 1 hours of birth (Table 2).

Table 2: Neonate bacterial sepsis versus neonate-care related variables at NICU, AHMC, (n = 317)

Variable	Confirmed neonatal sepsis		
	Yes (n/%)	No (n/%)	Total (n/%)
Neonate age (days)			
≤ 3	84(25.5)	129(40.7)	213 (67.2)
4-7	46(14.5)	28(8.8)	74(23.3)
8-28	17(5.4)	13(4.1)	30(9.5)
Neonates sex			
Male	82(25.9)	120(37.8)	202(63.7)
Female	65(20.5)	50(15.8)	115(36.3)
Gestational age (weeks)			
Preterm (<37)	57(18.0)	40(12.6)	97(30.6)
Term and postterm (≥37)	90(28.4)	130(41.0)	220(69.4)
Multiple birth			
Yes	17(5.4)	8(2.5)	25(7.9)
No	130(41.0)	162(51.1)	292(92.1)
Birth weight (kg)			
LBW (< 2.5)	100(31.5)	69(21.8)	169(53.3)
NBW and overweight (≥2.5)	47(14.8)	101(31.9)	148(46.7)
Place of delivery			
Health facility	145(45.7)	162(51.1)	307(96.8)
Home	2(0.06)	8(2.5)	10(3.2)
Mode of delivery			
SVD	79(24.9)	123(38.8)	202(63.7)
Instrumental	5(1.6)	3(0.9)	8(2.5)
CS	63(19.9)	44(13.9)	107(33.8)
Resuscitation at birth			
Yes	47(14.8)	18(5.7)	65(20.5)
No	100(31.5)	152(48.0)	252(79.5)
Duration of admission before sampling(hrs)			
≤48	140(44.2)	163(51.4)	303(95.6)
>48	7(2.2)	7(2.2)	14(4.4)
Invasive procedures			
Not at all	17(5.4)	89(28.1)	106(33.4)
Interavenous fluids	125(39.4)	77(24.3)	202(63.7)
Surgery	5(1.6)	4(1.3)	9(2.9)
Breast feeding initiation(hrs)			
< 1hrs	69(21.8)	98(30.9)	167(52.7)
1-2hrs	22(6.9)	25(7.9)	47(14.8)

Not at all	55(17.4)	48(15.1)	103(32.5)

Key: SVD-Spontaneous vaginal delivery; CS-Caesarian section; LBW-Low birth weight; NBW-Normal birth weight

Magnitude of neonatal sepsis and bacterial isolates

A total of 147 (46.4%) with a 95% Cl, 41.8 – 52.8%) of neonates had blood culture-confirmed bacterial sepsis (Figure 1). One of 147 neonates with confirmed bacterial sepsis has mixed bacterial infections. Three (2%) of positive blood cultures showed yeast cells. Gram-negative bacterial species account 107 (69.9%) and K. pneumoniae is the predominant isolates accounting for 72 (47.1%) of all isolates. Coagulase negative staphylococci (CoNS) 23 (15%) was the second most common isolate, followed by E. cloacae 18 (11.8%), A. baumannii 10 (6.5%), E. coli 7 (4.6%), S. aureus 8 (5.2%), Enterococcus spp. 10 (6.5%) and yeast cells 5(3.3%). The predominant organisms during the first 72 hours of life were gram-negatives 70 (80.5%) of the 87 isolate (Figure 1, 2 and Table 3).

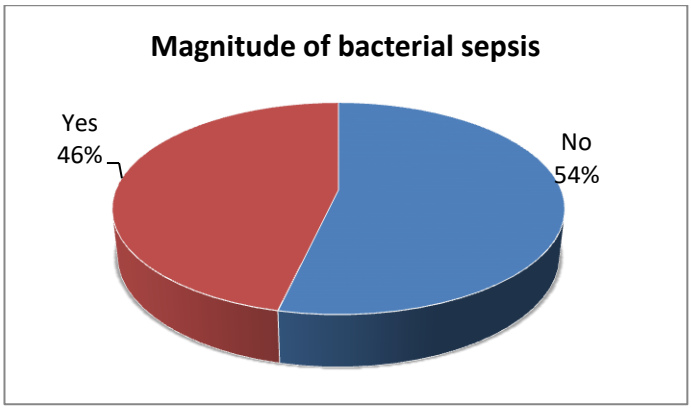

Figure 1: Magnitude of bacterial sepsis among neonates admitted to NICU AHMC, (n = 317)

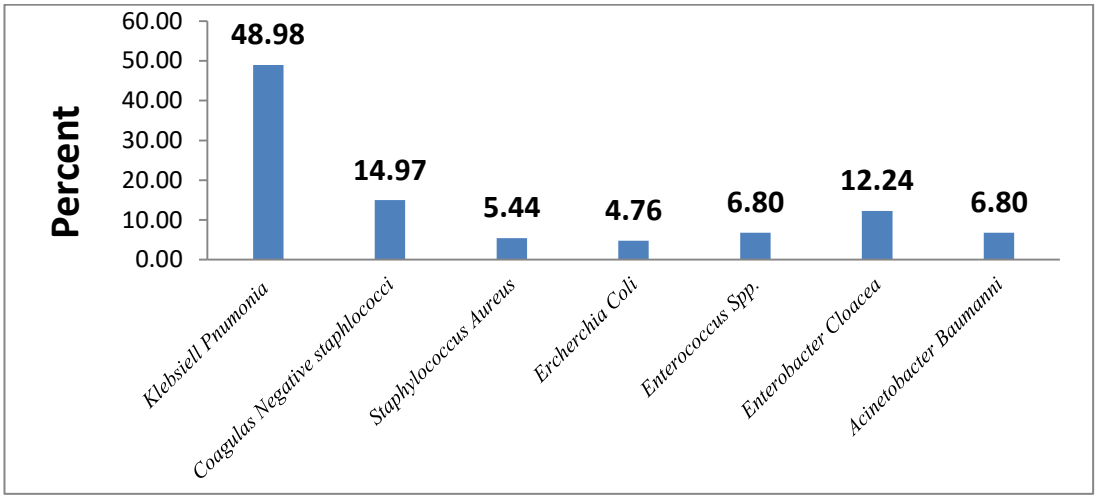

Figure 2: Bacterial isolates of blood culture from suspected neonates admitted at NICU, AHMC, (N = 147)

Table 3: Distribution of bacteria isolated from neonates based on their age at admission at NICU, AHMC, (N = 147)

Bcterial Isolates	Age of neonates at the times of admission		
	≤ 3 days	4-7 days	8-28 days
K. pneumonia	59(81.94%)	13(18.06%)	0
E. cloacae	4(22.22%)	9(50.00%)	5(27.78%)
A. baumannii	4(40.00%)	4(40.00%)	2(20.00%)
E. coli	3(42.86%)	2(28.57%)	2(28.57%)

CoNS	5(22.73%)	12(54.55%)	5(22.73%)
S. aurous	4(50.00%)	3(37.50%)	1(12.5%)
Enterococcus spp.	6(60.00%)	2(20.00%)	2(20.00%)

Key: CoNS – Couagulase negative staphylococcus

Factors associated with culture confirmed neonatal sepsis analysis

In bivariate logistic regression analysis age of neonates, neonates' sex, low birth weight, gestational age, multiple births, invasive procedure, mode of delivery, resuscitation at birth, amniotic fluid foul-smelling, history of maternal UTI/STI Infection, maternal fever, the onset of labor and duration of PRoM were statistically significant with culture-confirmed neonatal sepsis. After controlling for potential confounders in the multivariate model, the age of neonates, low birth weight, gestational age <37 weeks (preterm), invasive procedure (intravenous fluids) resuscitation at birth, and history of maternal UTI/STI were significantly associated with culture-confirmed neonatal sepsis.

There are maternal, neonatal and health related factors that have significant association with bacterial neonatal sepsis. The neonatal factors includes neonatal age, birth weight and gestational age; odds of being infected is 4.23(AOR= 4.23, 95% CI: 1.91, 9.65) and 6.07 (AOR= 6.07, 95% CI: 2.01, 18.22) higher among neonates of 4-7 days and 8-28 days old respectively compared to neonate age less than or equals to 3 days old. The odds of having neonatal bacterial sepsis is 2.24 (AOR=2.24, 95% CI: 1.1.06, 4.71) times higher among neonates having low birth weight compared to their counterpart and is 2.53 (AOR= 2.53, 95% CI: 1.19, 5.38). The maternal factors includes UTI/STIs and onset of labor. The odds of having being infected by bacteria is 3.73 (AOR= 3.73, 95% CI: 1.35, 10.28) and the odds of bacterial neonatal sepsis is 2.70 (AOR=2.70, 95% CI: 2.34, 25.41) times higher among neonate born by induced labor compared to neonates born by spantonous vaginal delivery.

Regarding health care related factors, factors like invasive procedure and resuscitation at birth shows significant association. The odd of being infected by bacteria is 5.21 (AOR= 5.21, 95% CI: 2.58, 10.51) times higher among neonate treated with IV fluids compared to neonate not treated with IV fluids and the odds of having neonatal bacterial sepsisis 7.85 (AOR= 7.85, 95% CI: 3.50, 17.58) times higher among neonates who are resuscitated at birth compared to non resuscitated neonates (Table 4).

Table 4: Bi-variable and multivariable logistic regression analyses of factors associated with culture confirmed neonatal sepsis among neonate admitted in NICU, AHMC,.(n=317)

Variables	Confirmed neonatal sepsis		COR(95%Cl)	AOR(95%Cl)
	Yes	No		
Maternal characteristics				
Duration of PROM (hrs)				
Not ruptured	11(55%)	9(45%)	1.61 (.64, 4.04)	0.19(.03, 1.34)
≥ 18	36(55.4%)	29(44.6%)	1.64(.94, 2.85)*	0.55(.22, 1.37)
<18	100(43.1%)	132(56.9%)	1	1
Onset of Labor				
Spontaneous	113(40.8%)	164(59.2%)	1	1
Induced	34(85.0%)	6(15.0%)	2.88(1.08, 7.69)*	2.70(2.34, 25.41)**
Presence of Fever				
Yes	43(70.5%)	18(29.5%)	3.49(1.91, 6.39)*	1.53(.58, 4.04)
No	104(40.6%)	152(59.4%)	1	1
UTI/STI Infection				
Yes	42(79.2%)	11(20.8%)	5.78(2.85, 11.74)*	3.73(1.35, 10.28)**
No	105(39.8%)	159(60.2%)	1	
Amniotic fluid foul-smelling				
Yes	30(62.5%)	18(37.5%)	2.17 (1.15, 4.07)*	1.29(.55, 3.02)
No	117(43.5%)	152(56.5%)	1	1
Neonates characteristics				
Neonates Sex				

Male	82(40.6%)	120(59.4%)	1	1
Female	65(56.5%)	50(43.5%)	1.90 (1.20, 3.02)*	2.34(.21, 4.51)
Neonate age(day)				
≤ 3 days	9(40.9%)	13(59.1%)	1	1
4-7 days	118(47.8%)	129(52.2%)	2.52(1.46, 4.35)*	4.30 (1.91, 9.65)**
8-28 days	20(41.7%)	28(58.3%)	2.01(.93, 4.35)*	6.07(2.02, 18.22)**
Birth weight				
LBW	100(59.2%)	69(40.8%)	3.11 (1.96, 4.95)*	2.24 (1.06, 4.71)**
NBW+	47(31.8%)	101(68.2%)	1	1
Gestation age				
Preterm	57(58.8%)	40(41.2%)	1.73 (1.06, 2.82)*	2.53(1.19, 5.38)**
Term+	90(40.9%)	130(59.1%)	1	1
Multiple birth				
Yes	17(68.0%)	8(32.0%)	2.65 (1.11, 6.33)*	1.22(.34, 4.38)
No	130(44.5%)	162(55.5%)	1	1
Healthcare related characteristics				
Mode Of Delivery				
SVD	79(39.1%)	123(60.9%)	1	1
Instrumental	5(62.5%)	3(37.5%)	2.60 (.60, 11.16)	3.09(.51, 18.54)
C/S	63(58.9%)	44(41.1%)	2.23 (1.38, 3.59)*	1.19(.55, 2.57)
Invasive Procedure				
Not at all	17(16.0%)	89(84.0%)	1	1
Intravenous fluids	125(61.9%)	77(38.1%)	8.03(4.49, 14.35)*	5.21(2.58, 10.51)**
Surgery	4(50.0%)	4(50.0%)	4.94(1.13, 21.63)*	4.07(.69, 23.93)
Resuscitation at birth				
Yes	47(72.3%)	18(27.7%)	3.97 (2.18, 7.23)*	7.85(3.50, 17.58)**
No	100(39.7%)	152(60.3%)	1	1

Key: SVD-Spontaneous vaginal delivery; C/S-Caesarian-Section; LBW-Low birth weight; NBW-Normal birth weight

Discussion

Bacterial neonatal sepsis remains a major clinical problem of neonatology with high morbidity and mortality rate, especially in developing countries including Ethiopia. The overall culture positivity rate of bacterial isolates identified from neonates with symptoms of neonatal sepsis in this study was 47.3%, almost similar with the results of study conducted in Ethiopia, Gondar (46.6%) but lower than Sudan (61.3%) and Yemen (57%) (12,32) and slightly higher than the studies conducted in Nigeria (43.5%), Egypt (42.7%) and and Addis Ababa (44.7%) (1,14,15,33) and higher than the study done in Nepal (16.9%), Tanzania (33%), Ghana (17.3%) and Ethiopia; Asella (29.4%), Gondar (32.1%) and Dire Dawa (32.1%) (3,13,19,34-36). This variations of the study could be due to methodological variation, and the difference in the study setting (place of delivery, mode of delivery and type of invasive procedure), which might affect the culture positivity rate, and again, it may be due to differences in predisposing factors and infection control practices in the delivery service providing and treatment centers.

In our study, gram-negative and gram-positive bacterias represented 69.9% and 26.8%, respectively, of all isolates, and gram negative were predominant in both EOS and LOS. In agreement with our results, many studies reported the predominance of gram-negative organisms in different areas of the developing world (4). These results are consistent with studies conducted in Nepal, Pakistan, Egypt, and in Ethiopia, particulalry in Addis Ababa and Dire Dawa (13,15,33,37). The predominance of gram negative organisms may be due to the indiscriminate and inappropriate use of antibiotics (alter gut colonization), lack of hygienic practices at the place of delivery, poor cord care, and unhygienic newborn care practices and high chance of acquiring vaginal gram-negative bacteria among neonates (38). On the contrary, other

studies from India, Tehran, Ghana, Nigeria, and Ethiopia (Gondar and Asella) reported a predominance of gram-positive organisms (1,3,19,36,39,40). The causative organism varies due to the geographical area. This may be due to difference in latitude, which may favor or unfavor the distribution or growth of bacteria. Proximity to equator could be the cause for bacterial variability (41). Since Ethiopia (Adama town) is proximate to equator than Egypt, Pakistan and Nepal this may be difference in bacterial cause of neonatal sepsis.

This study showed that resuscitation at birth is the most important neonatal factor predisposing to sepsis. Newborns who had resuscitation shows significant association with neonatal bacterial sepsis. The result of this study is similarly with study done in Tanzania by Jabiri, Wella et al and Eastern Ethiopia by Seyoum, Alemayehu et al (13,35) On the contrary, a study done in Mekelle (25) revealed insignificant association. This discrepancy may be explained by the differences in ventilation procedure safety practice and study setting and infection prevention measures and cleanliness of material for resuscitation and the level of application of infection prevention procedures among health care providers.

Age of neonates is another associated factor for bacterial neonatal sepsis in this study and studies done elsewhere (13,42). This could be due to immature immunity of the neonate that make them vulnerable to hospital-acquired infection (more than 3 days).

Neonates born to mothers who had a UTI/STI during the index pregnancy were nearly four times more likely to develop confirmed sepsis than those neonates born to mothers who did not have a UTI/STI during the index pregnancy. This finding is supported by the study conducted in Ghana (23) and Ethiopia; in Bishoftu, Jinka, and Mekelle (25,43,44). This might be due to late diagnosis and treating of UTI that could result in colonization of the birth canal by bacteria and neonate acquire the bacteria during the birth process from birth canal.

This study revealed that low birth weight and prematurity are long established neonatal factors predisposing to sepsis. The lower the birth weight (less than 2.5kg) and prematurity, the greater the vulnerability of the newborn to septicemia by 2.23 and 2.53 folds respectively, as such neonates were expected to be more debilitated, the weight could affect the immune status and their immunity could be immature (45). But the study done in Nepal and Bishoftu, Ethiopia indicates that being preterm was not significantly associated with the occurrence of neonatal sepsis (43,46). The possible justifications for this contradiction may be due to geographic location, and its risk factor, or it may be due to the fact that neonate with low birth weight and premature maynot have strongly developed immunity at the time of their birth.

This study also indicates that intravenous fluids medication is the most important healthcare-related factors contributed to sepsis. Neonates, who received intravenous fluids medication, were 5.21 times more likely to develop sepsis than those neonates who did not receive the medication. This is consistent with studies conducted in southeastern Mexico, Brazil and Egypt (47-49). In this case, the cause of infection may not be intravenous fluid,, but it may be the sterility techniques the health care providers used. Therefore, the level of application of infection prevention measures varies among health care providers and accessibility of infection prevention materials in health institutions. Levels of health care providers adherence to aseptic techniques determines infection due to Intravenous treatment. Intravenous liquids are mixed and administered by nurses, sometimes under suboptimal infection control conditions. This may be due to health care providers not strictly adhering to aseptic techniques or procedures.

Since the study was done on admitted neonates, results might lack generalizability to the entire population in the catchment area. It was not possible to identify anaerobic blood culture due to financial constraints, where the research was conducted. Since the study is done on admitted neonates and data was not collected from the community, thus results might lack generalizability to the entire population in the catchment area.

Conclusion and recommendations

Nearly half (47.3%) of the neonate developed bacterial neonatal sepsis which is the significant public health problem among the population with major contributing factors like birth weight, intravenous fluid medication, resuscitation at birth and history of maternal UTI/STI. So, strictly adhere to infection prevention control during services like IV fluid provision and resuscitation and UTIs/STIs screening early treatement for all pregnant women during antenatal care and followup were recommended to reduce bacterial neonatal sepsis.

Health care providers should strictly adhere to infection prevention and aseptic techniques procedure during services like IV fluid provision and resuscitation and should give more attention for prevention of sepsis among preterm neonate and neonate of age 4 to 28 days since preterm neonates are susceptible for infections. HCPs should be concerned about UTIs/STIs screening for all pregnant women during antenatal care and followup and they should adhere to aseptic techniques during lobor induction.

ACRONYMS/ABBREVIATION

AHMC	Adama Hospital Medical College
AOR	Adjusted Odd Ratio
ATCC	American Type Culture Collection
APGAR	Activity, Pulse, Grimace, Apperance, and Respiration
CS	Caesarean Section
CoNS	Coagulase Negative Staphylococci
CLSI	Clinical and Laboratory Standard Institute
EDHS	Ethiopia Demographic and Health Survey
EOS	Early Onset Sepsis
ESBL	Extended-Spectrum Beta-Lactamase
GBS	Group B Streptococcus

HIV	Human Immunodeficiency Virus
LOS	Late-Onset Sepsis
MAC	MAcConkey
MDG	Millennium Development Goal
MDR	Multi-Drug Resistant
MHA	Mueller Hinton Agar
MRSA	Methicillin Resistant Staphylococcus Aureus
NICU	Neonatal Intensive Care Unit
PRoM	Prolonged Rupture of Membranes
SD	Standard Deviation
SDG	Sustainable Development Goal
SPSS	Statistical Package for the Social Science
STI	Sexually Transmitted Infection
UTI	Urinary Tract Infection

Ethics approval and consent to participate

Ethical clearance was obtained from Adama Hospital Medical College Ethical Review Board(Ref. No, 0382/K-373/12) and the Oromia Administration Health Bureau support letter (Ref.No, BEFO/HBTFU/1-16/353) was communicated to AHMC and the center where the pre-test was performed. Informed consent was obtained from each parent/guard before the initiation of data collection.

Consent for publication

Not Applicable

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request**.**

Competing Interest

The authors declare that we do not have any competing interests

Funding

Not applicable

Authors` contribution

All authors, TO, TS and EM contributed into concept preparation and design, acquisition of data, analysis and interpretation of data and writing the final manuscript. All authors read and approve the final manuscript for publication.

Acknowledgment

The authors want acknowledge Adama Hospital Medical College(AHMC), the mothers of neonate as well as the family for their volunteer ness of participating in this research and allowing as to take blood sample of the neonate. Finally we acknowledge the staffs of AHMC working in NICU ward for their support in withdrawing blood sample from the neonate and collecting data from mother.

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