**ORIGINAL ARTICLE**

# BACTERIAL PROFILE AND THEIR ANTIBIOTIC RESISTANCE PATTERNS FROM BLOOD CULTURE AND ASSOCIATED RISK FACTORS IN INTENSIVE CARE UNIT AT THE UNIVERSITY OF GONDAR COMPREHENSIVE SPECIALIZED HOSPITAL, NORTHWEST ETHIOPIA

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## ABSTRACT

***Background:*** *The burden of blood stream infection and antibiotic resistance in intensive care unit (ICU) is higher when compared with other settings. The data on the infection rate and changing antibiotic resistance trends are important for infection control activities. The aim of this study was to assess the magnitude and risk factors of bacterial pathogens and their antibiotic resistance patterns of blood culture isolates from ICU patients at the University of Gondar Comprehensive Specialized Hospital, Northwest Ethiopia.*

***Materials and Methods:*** *A cross-sectional study was conducted from February to May 2018. Blood specimen was collected aseptically from 384 patients and inoculated on appropriate culture media. Identification of bacterial pathogens and antibiotic susceptibility tests were performed using bacteriological standard methods. Presence of Extended Spectrum Beta Lactamase (ESBL) enzymes was confirmed by combined disk diffusion method. Cefoxitin was tested as a surrogate marker for oxacillin (methicillin) to detect methicillin resistant Staphylococcus aureus (MRSA). All materials, equipment, antibiotic disks and procedures were adequately controlled. Data were analyzed using SPSS version 20 by applying binary logistic regression model with multivariable analysis. P-value <0.05 at 95% CI was considered as statistically significant.*

***Results:*** *Of the 384 study participants, 53.9% were neonates and the most common reasons (40.9%) for hospital admission was early onset neonatal sepsis. From the total samples, 25% were culture positive. K.pneumoniae was the predominant isolate both in neonatal and pediatric ICU and E.coli in adult ICU. Majority of Gram positive and Gram negative isolates were highly sensitive to Clindamycin (83%) and meropenem (84.8%) & ciprofloxacin(83.5%) respectively. The prevalence of MRSA and ESBL enzyme producers was 57.1% and 78.8%, respectively. Age and sex were significantly associated factors with blood stream infection.*

***Conclusion:*** *Klebsiella species were the predominant bacterial isolates in ICU settings. Clindamycin for Gram positive, meropenem and ciprofloxacin for Gram negative isolates were highly effective. Antibiotic resistance due to ESBL enzyme production is alarmingly high. The high prevalence of Klebseilla species in the ICU may signal inappropriate use of antibiotics and poor infection prevention control practice in these settings. Being male and neonate were more likely to develop bacterial bloodstream infection.*

***Key words:*** *Antibiotic resistance, ICU, Blood Culture, University of Gondar Comprehensive Specialized Hospital, Ethiopia.*

## INTRODUCTION

Intensive care units (ICUs) are the most life-saving units for critically ill patients in a hospital that are evaluated and followed by highly specialized clinicians and nurses(1).Most of the conditions that are treated at ICU are life-threatening, severe injuries and diseases that require constant monitoring and support from specialized equipment and medications to ensure normal body functions(2).

Blood stream infection (BSI) is a leading cause of death in the ICU as high as 60% and twice as much in those patients with a nosocomial infection (3). Intensive care units are one of the potential sources

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of nosocomial infections even in developed countries with extensive infection control measures(4). But in developing countries, the prevalence of ICU-acquired infections is substantially higher than in industrialized countries. The most frequently reported bacterial pathogens in ICU settings are *Staphylococcus aureus, Enterobacteriaceae, Pseudomonas* species, *Enterococcus* species*, Candida* species, Coagulase-negative staphylococci (CoNS) and *Acineto bacter* species(5-7).

In the ICUs, the rate of infections with antibiotic-resistant bacteria is increasing, primarily due to increased use of invasive procedures and specialized devices, overuse of antibiotics and misuse due to incorrect diagnosis, excessive variations of the antibiotic market, and frequent use due to either incorrect prescription or inadequate enforcement (3, 8, 9).Failures to identify the existence of antibiotic resistance are also important factors facilitating the spread of antibiotic resistance due to inadequate infection control practices(10, 11).

The mortality and morbidity rates of BSIs in ICU are higher due to the limited available resources associated with the quality of ICU's health care system (12, 13). There is, however, a lack of available data in developing countries such as Ethiopia. Therefore, the overall aim of this study was to assess the magnitude of bacterial isolates, associated risk factors and their antibiotic resistance patterns from blood specimens in the ICU at the University of Gondar Comprehensive Specialized Hospital.

## MATERIALS AND METHODS

**Study design, area and period:** A cross-sectional study was conducted at the University of Gondar Comprehensive Specialized Hospital from February to May 2018. The hospital is a teaching hospital which is located in Gondar town, 750 km far from Addis Ababa in the Northwest Ethiopia. In this town, there are 8 health centers, 21 private clinics and one hospital. The hospital provides surgical, medical, pediatric, gynecologic, obstetric, oncologic and ophthalmologic services to the community for over 5 million inhabitants. The hospital has only 4 beds for adult ICU (AICU),6 beds for pediatric ICU (PICU) and 36 beds for neonatal ICU (NICU)(14).

**Population**

**Source population:** were all patients who were admitted at the University of Gondar Comprehensive Specialized Hospital during the study period.

**Study population:** were all patients who were admitted to ICUs and suggested of developing bacterial blood stream infection at the University of Gondar Comprehensive Specialized Hospital.

**Inclusion and Exclusion criteria**

**Inclusion criteria:** All patients who were admitted to AICU, PICU and NICU due to bloodstream infection during the study period were included in the study.

**Exclusion criteria:** Patients who were admitted to ICUs due to other than bloodstream infection and patients unable to give specimen for culture were excluded from the study.

**Variables**

**Dependent variables:** Bacterial isolates and antibiotic resistance pattern were the outcome variables.

**Independent variables:** Age, sex, educational background, occupation, residence, patient settings, rea-

son for ICU admission, antibiotic taken, devices used during admission were independent variables.

**Sample size and sampling technique**

Using convenient sampling technique, a total of 384 septicaemia suspected ICU patients were enrolled.

**Operational Definitions**

**EONS:** refers to sepsis presenting in the first 7 days of life

**LONS:** refers to presentation of sepsis after 7 days.

**Neonate:** is a child under 28 days of age

**Paediatric:** A child who are known to be less than 18 years of age

**Adult:** An individual beyond 18 years of *age*

**Data collection methods**

Socio demographic data were collected using structured questionnaire. From each patient, two bottles of blood samples from different sites were collected by trained nurse working in the ICU under the supervision of the investigator considering all aseptic techniques. For all samples, phlebotomy was performed after disinfection of vein puncture site with 2% tincture iodine followed by 70% alcohol. Ten ml, 5ml and 1ml of blood samples per bottle were collected from adults, pediatrics, and neonates, respectively which were then inoculated in to home made tryptic soya broth (15).

**Laboratory processing**

After inoculation, the blood samples were incubated immediately at 35-37$^0$C aerobically based on the standard operating procedure of manual blood culturing technique (University of Gondar Comprehensive Specialized Hospital laboratory blood culturing manual). After 18 hours of incubation, Gram stain was done and sub cultured on to 5% sheep blood agar, Mac Conkey agar (Oxoid) and Chocolate agar (Oxoid). Blood agar and Chocolate agar plates were incubated at 35-37$^o$C in 5% $CO_2$ and MacConkey agar without $CO_2$ for 18-24 hours. If there was no growth on the blood agar and chocolate agar, they were further incubated for additional 24 hours. If still no growth, Chocolate agar only was incubated for further 24 hours. The bottles were incubated for up to seven days under observation and if no growth reported as negative.

Presumptive identification of bacteria was performed based on sign of growth on blood culture bottle, Gram reaction, and colony characteristics. Presumptively isolated pathogens were further identified to species level using different biochemical tests. Catalase, coagulase, bacitracin, and optochin tests were performed for Gram positive bacterial pathogens; triple sugar iron agar, indole, citrate utilization, urease production, lysine decarboxylase, and motility test were performed for Gram negative bacteria.

**Antibiotic susceptibility testing**

Once species were identified, antibiotic **s**usceptibility testing was performed for different antibiotics using the modified Kirby-Bauer disk diffusion method by using Muller Hinton Agar (Oxoid), Chocolate agar (Oxoid) and Muller Hinton agar with 5% sheep blood (Oxoid) based on the type of bacteria isolated. The suspension of the test organism was prepared by picking parts of similar test organisms with a sterile wire loop and suspend in test tube containing sterile physiological saline. The density of the suspension was determined using 0.5 McFarland standards. Using sterile cotton swab test organism was introduced over the entire surface of the agar. Antibiotic discs were distributed 15mm away from the edge and ≥24mm apart from each other and incubated at 37$^o$C for 24 hours. The results were interpreted as suscepti-

ble, intermediate or resistant based on the 2017 Clinical and Laboratory Standards Institute (CLSI) guideline(16). Cefoxitin (30µg) was tested as a surrogate marker for oxacillin (methicillin) to detect methicillin resistant *Staphylococcus aureus* (MRSA). Presence of extended spectrum beta lactamase (ESBL) for *Enterobacteriaceae* was also suggested by screening resistance to a third generation cephalosporins such as cefotaxime (30µg), ceftriaxone (30µg) or ceftazidime (30µg) according to the CLSI screening criteria. A combined disk method was used as a confirmatory phenotypic method for ESBL detection. Ceftazidime (30µg) and cefotaxime (30µg) disks alone and their combinations with clavulanate (30µg/10µg) were used for phenotypic confirmations of ESBL production(16).

**Data quality control**

The reliability of the study findings was guaranteed by implementing quality control measures throughout the whole process of the laboratory work. All materials, equipment, antibiotic disks and procedures were adequately controlled. Culture media were tested for sterility and performance. Pre-analytical, analytical and post-analytical stages of quality assurance that are incorporated in standard operating procedures of the University of Gondar Comprehensive Specialized Hospital Microbiology laboratory were strictly followed. Control strains of *S.aureus* ATCC 25923*, E.coli* ATCC 25922 and *P.aeruginosa* ATCC 27853 were used to confirm consistency of materials, methods, and results.

**Data analysis and interpretation**

Data were entered, coded and cleaned using Epi-info version 7.0 statistical software and then transferred to SPSS software version 20 for further data processing and analysis. Text descriptions, tables, and figures were used to describe the relevant findings of the study. The crude odds ratios with a 95% confidence interval were estimated in the binary logistic regression analysis to assess the association between each independent variable and the outcome variable. Those independent variables with a p - values of less than or equal to 0.20 were tested in the multivariate logistic regression to get the adjusted effect of each covariate. P-value ≤0.05 were considered as statistically significant.

**Ethics approval and consent to participate**

This study protocol was approved by the ethical review committee of the School of Biomedical and Laboratory Sciences, College of Medicine and Health Sciences, University of Gondar. The consent and/or assent forms were taken from caretakers and/or study participants after explaining the objective of the study.

## RESULTS

**Socio-demographic characteristics of study participants**

From the 384 septicemia suspected ICU patients, 205 (53.4%) of the study participants were males and 179 (46.6%) were females. The median age of the participants' was 21 days. Majority of the study participants were Neonates 207(53.9%) and rural dwellers, 267 (69.5%) (Table 1).

**Clinical information of study participants**

The most common reasons for hospital admission include early onset neonatal sepsis 157 (40.9%) followed by pyogenic meningitis 55(14.3%) and late onset neonatal sepsis, 38(9.9%). From the total patients, 231(60.3%) were referred from other wards. In relation to antibiotic usage, 167(43.5%) had taken

antibiotics before sample collection and 217(56.5%) didn't take any antibiotics. Most commonly prescribed antibiotics for those patients were ampicillin plus gentamicin, 88(22.9%) and metronidazole plus ceftriaxone, 27 (7%). Different mechanical devices were used for 172(44.8%) of the study participants

**Table 1:** Socio-demographic characteristics and clinical information of ICU patients at the University of Gondar Comprehensive Specialized Hospital from February to May 2018.

| | **Independent Variables (n=384)** | **Number** | **%** |
|---|---|---|---|
| **Age** | Neonates (0 -28 days) | 207 | 53.9 |
| | Pediatrics (29 day-18 years) | 113 | 29.4 |
| | Adults (>18 years) | 64 | 16.7 |
| **Sex** | Male | 205 | 53.4 |
| | Female | 179 | 46.6 |
| **Residence** | Rural | 267 | 69.5 |
| | Urban | 117 | 30.5 |
| **Occupation** | Farmer | 19 | 4.9 |
| | Civil servants | 6 | 1.6 |
| | Self-employee | 20 | 5.2 |
| | Daily laborer | 4 | 1.0 |
| | House wife | 17 | 4.4 |
| | Merchant | 2 | 0.5 |
| | Others* | 2 | 0.5 |
| | N/A | 314 | 81.8 |
| **Educational status** | Illiterate | 17 | 4.4 |
| | Primary | 58 | 15.1 |
| | Secondary | 28 | 7.3 |
| | College and above | 6 | 1.6 |
| | N/A* | 275 | 71.6 |
| **Clinical diagnosis** | EONS | 157 | 40.9 |
| | LONS | 38 | 9.9 |
| | Pyogenic meningitis | 55 | 14.3 |
| | HAS | 29 | 7.6 |
| | Localized infection | 26 | 6.8 |
| | Pneumonia | 37 | 9.6 |
| | Complicated UTI | 11 | 2.9 |
| | Infective endocarditis | 21 | 5.5 |
| | Others** | 10 | 2.6 |
| **Admitted from** | Other wards | 231 | 60.2 |
| | Health center | 59 | 15.4 |
| | Other hospital | 23 | 6.0 |
| | Home | 71 | 18.5 |
| **Prior antibiotic therapy** | Yes | 167 | 43.5 |
| | No | 217 | 56.5 |
| **Mechanical device used** | Yes | 172 | 44.8 |
| | No | 212 | 55.2 |

***Keys**: N/A* - Patients who are <6-years-old, others* - Carpenter, Beiger, Others**- multiple trauma, hemorrhagic shock and acute kidney injury, EONS- Early Onset Neonatal Sepsis, LONS-Late Onset Neonatal Sepsis, HAS- Hospital Acquired Sepsis, UTI-Urinary Tract Infection, NICU-Neonatal Intensive Care Unit, PICU- Paediatric Intensive Care Unit, AICU-Adult Intensive Care Unit.*

**Bacterial profile**

From the 384 blood samples, 96 (25%) were culture positive. Of these, 67 (69.8%) of bacterial isolates were Gram-negative, while the remaining 29(30.2%) were Gram positive. Nearly one-third, 65/207 (31.4%) of the bacterial isolates were isolated from blood samples of neonates, followed by pediatrics, 20/113(17.7%). *Klebsseilla* species specifically *K.pneumoniae* was the predominant isolate both in neonatal ICU and pediatric ICU and *E.coli* in adult ICU (Table 2).

**Table 2:** Distribution of bacterial isolates of blood culture in different ICU patients at the University of Gondar Comprehensive Specialized Hospital from February to May 2018.

| Type of organism isolated | NICU (207) n (%) | PICU (113) n (%) | AICU (64) n (%) | Total (384) n(%) |
|---|---|---|---|---|
| ***K.pneumoniae*** | 10 (4.8) | 6 (5.3) | 2 (3.1) | 18 (4.7) |
| **CoNS** | 8 (3.9) | 4 (3.5) | 1 (1.5) | 13 (3.4) |
| ***K.ozenae*** | 8 (3.9) | 0 | 2(3.1) | 10 (2.6) |
| ***K.rhinose*** | 7(3.4) | 1 (0.9) | 0 | 8 (2.1) |
| ***E.coli*** | 3 (1.4) | 1 (0.9) | 3 (4.7) | 7 (1.8) |
| ***Citrobacter* species** | 6 (2.9) | 0 | 1 (1.5) | 7 (1.8) |
| ***E.cloacae*** | 7 (3.4) | 0 | 0 | 7 (1.8) |
| ***S.aureus*** | 4 (1.9) | 3 (2.6) | 0 | 7 (1.8) |
| ***S.viridans*** | 4 (1.9) | 1 (0.9) | 1 (1.5) | 6 (1.6) |
| ***Providencia* species** | 3 (1.4) | 1 (0.9) | 1 (1.5) | 5 (1.3) |
| ***Serratia* species** | 4 (1.9) | 0 | 0 | 4 (1) |
| ***S.pneumonae*** | 1 (0.5) | 2 (1.8) | 0 | 3 (0.8) |
| ***Neisseria* species** | 0 | 1 (0.9) | 0 | 1 (0.3) |
| **Total** | 65(31.4) | 20(17.7) | 11(17.2) | 96 (25) |

***Keys:*** *NICU-Neonatal Intensive Care Unit, PICU- Paediatric Intensive Care Unit, AICU-Adult Intensive Care Unit.*

**Antibiotic susceptibility patterns of Gram positive and Gram negative isolates**

Majority of the Gram positive bacterial isolates were found to be resistant to tetracycline 27(93.1%), followed byt rimethoprim-sulfamethoxazole 26(89.6%), erythromycin 26(89.6 %), and chloramphenicol 21 (72.4%). CoNS (100%) were also resistant to penicillin. On the other hand, majority 24(82.7%) of the Gram positive isolates were sensitive to clindamycin (Table 3).

Most of the Gram negative isolates were highly resistant to trimethoprim-sulfamethoxazole 64(95.5%) followed by chloramphenicol 57(85.6%) and ceftriaxone 51(76%) and least resistant to meropenem 9/66(13.6%) and ciprofloxacin 9/67(13.4%). *K. pneumoniae* demonstrated high level of resistance to amoxicillin/clavulanate 17(94.4%), ceftriaxone 17 (94.4%), chloramphenicol15(83.3%), trimethoprim-sulfamethoxazole 16(88.9%) and gentamicin 9(50%). On the other hand, meropenem16 (88.9%) and ciprofloxacin 13(72.2%) were effective against this isolate (Table 3).

**Table 3:** Antibiotic susceptibility patterns of bacterial isolates from blood culture of ICU patients at the University of Gondar Comprehensive Specialized Hospital from February to May 2018.

| Bacterial isolate | Patterns | PG N (%) | FOX N (%) | CIP N (%) | GN N (%) | ERY N (%) | CAF N (%) | SXT N (%) | CRO N (%) | DA N (%) | TE N (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Gram positive** | | | | | | | | | | | |
| CoNS **n=13** | S | 0 (0) | 5 (38.5) | 8 (61.5) | 6 (46.2) | 2 (15.4) | 5 (38.5) | 2 (15.4) | - | 12 (92.3) | 2(15.4) |
| | I | 0 (0) | 0 (0) | 1 (7.7) | 1 (7.7) | 0 (0) | 0 (0) | 0 (0) | - | 0 (0) | 0 (0) |
| | R | 13 (100) | 8 (61.5) | 4 (30.8) | 6 (46.2) | 11 (84.6) | 8 (61.5) | 11(84.6) | - | 1 (7.7) | 11(84.6) |
| ***S.aureus*** **n=7** | S | 1 (14.3) | 3(42.9) | 6 (85.7) | 4 (57.1) | 1 (14.3) | 3(42.9) | 1 (14.3) | - | 5 (71.4) | 0 (0) |
| | I | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | - | 0 (0) | 0 (0) |
| | R | 6 (85.7) | 4 (57.1) | 1 (14.3) | 3 (42.9) | 6 (85.7) | 4 (57.1) | 6 (85.7) | - | 2 (28.6) | 7 (100) |
| ***S.viridans*** **n=6** | S | - | - | - | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 5 (83.3) | 0 (0) |
| | I | - | - | - | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |
| | R | - | - | - | - | 6 (100) | 6 (100) | 6 (100) | 6 (100) | 1 (16.7) | 6(100) |
| ***S.pneumoniae*** **n=3** | S | - | - | - | - | 0 (0) | 0 (0) | 0 (0) | - | 2 (66.7) | 0 (0) |
| | I | - | - | - | - | 0 (0) | 0 (0) | 0 (0) | - | 0 (0) | 0 (0) |
| | R | - | - | - | - | 3 (100) | 3 (0) | 3 (100) | - | 1(33.3) | 3 (100) |

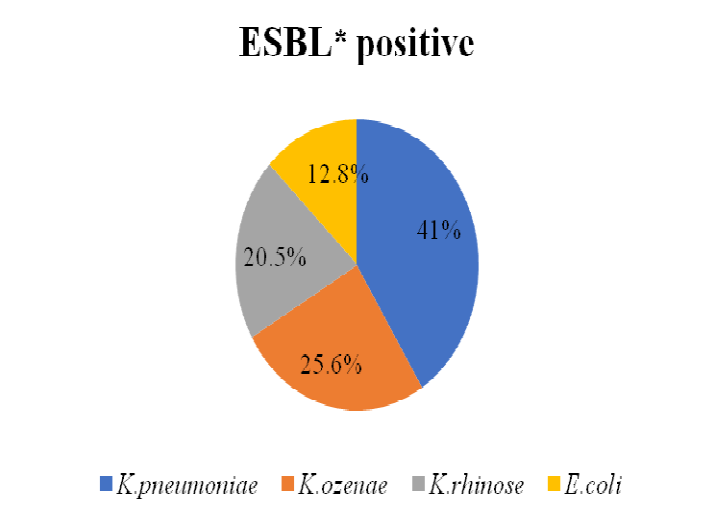
| Bacterial isolate | Pattern | AMP N (%) | Amox/cla N (%) | CIP N (%) | GN N (%) | CAF N (%) | SXT N (%) | CRO N (%) | CAZ N (%) | CTX N (%) | MER N (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Gram negative** | | | | | | | | | | | |
| ***K.pneumoniae*** **n=18** | S | - | 0 (0) | 13 (72.2) | 8(44.4) | 3(16.7) | 2 (11) | 1(5.5) | 1(5.5) | 6 (33) | 16 (88.9) |
| | I | - | 1(5.6) | 2 (11.1) | 1(5.60) | 0 (0) | 0 (0) | 0 (0) | 1(5.5) | 0 (0) | 0 (0) |
| | R | - | 17 (94.4) | 3 (16.7) | 9 (50) | 15(83.3) | 16(88.9) | 17(94.4) | 16 (89) | 12 (67) | 2(11.1) |
| ***K.ozanae*** **n=10** | S | - | 0 (0) | 10 (100) | 3 (30) | 2 (20) | 0 (0) | 2 (20) | 0 (0) | 3 (30) | 8 (80) |
| | I | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 1 (10) |
| | R | - | 10 (100) | 0 (0) | 7 (70) | 8 (80) | 10 (100) | 8 (80) | 10(100) | 7 (70) | 1 (10) |
| ***K.rhinose*** ***n*=8** | S | - | 0 (0) | 8 (100) | 2 (25) | 1 (12.5) | 0 (0) | 0 (0) | 3 (37.5) | 2 (25) | 6 (75) |
| | I | - | 0 (0) | 0 (0) | 0 (0) | 2 (25) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |
| | R | - | 8 (100) | 0 (0) | 6 (75) | 5 (62.5) | 8 (100) | 8 (100) | 5 (62.5) | 6 (75) | 2 (25) |
| ***E.coli*** **n=7** | S | 0 (0) | 2 (28.6) | 7 (100) | 6 (85.7) | 1(14.3) | 0 (0) | 1 (14.3) | 0 (0) | 0 (0) | 7 (100) |
| | I | 1 (14.3) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |
| | R | 6 (85.7) | 5 (71.4) | 0 (0) | 1(14.3) | 6 (85.7) | 7 (100) | 6 (85.7) | 7 (100) | 7 (100) | 0 (0) |
| ***E.cloacae*** **n=7** | S | - | - | 7 (100) | 2 (28.6) | 1 (14.3) | 1 (14) | 7 (00) | 7 (100) | 7 (100) | 7 (100) |
| | I | - | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |
| | R | - | - | 0 (0) | 5 (71.4) | 6 (85.6) | 6 (86) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |

| Bacterial isolate | Pattern | AMP N (%) | Amox/cla N (%) | CIP N (%) | GN N (%) | CAF N (%) | SXT N (%) | CRO N (%) | CAZ N (%) | CTX N (%) | MER N (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Gram negative** | | | | | | | | | | | |
| ***Citrobacter* species n=7** | S | - | - | 5 (71.4) | 0 (0) | 0 (0) | 0 (0) | 2(28.6) | 2(28.6) | 1(14.3) | 6(85.7) |
| | I | - | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |
| | R | - | - | 2 (28.6) | 7 (100) | 7 (100) | 7 (100) | 5 (71.4) | 5 (71.4) | 6 (85.7) | 1 (14.3) |
| ***Providencia* species n=5** | S | - | - | 3 (60) | 0 (0) | 0 (0) | 0 (0) | 2 (40) | 1 (20) | 1(20) | 3 (60) |
| | I | - | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) |
| | R | - | - | 2 (40) | 5 (100) | 5 (100) | 5 (100) | 3 (60) | 4 (80) | 4 (80) | 2 (40) |
| ***Serratia* *n*=4** | S | - | - | 2 (50) | 0 (0) | 0 (0) | 0 (0) | 1 (25) | 0 (0) | - | 3 (75) |
| | I | - | - | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | 0 (0) | - | 0 (0) |
| | R | - | - | 2 (50) | 4 (100) | 4 (100) | 4(100) | 3 (75) | 4 (100) | - | 1 (25) |
| ***Neisseria* species n=1** | S | - | 0 (0) | 1 (100) | - | 0 (0) | 0 (0) | 0 (0) | - | - | - |
| | I | - | 0 (0) | 0 (0) | - | 0 (0) | 0 (0) | 0 (0) | - | - | - |
| | R | - | 1 (100) | 0 (0) | - | 1(100) | 1(100) | 1 (100) | - | - | - |

***Keys:*** *S-Sensitive, I- Intermediate,R- Resistance. PG-Penicillin G,CIP- Ciprofloxacin, FOX-Cefoxitin, GN-Gentamicin, ERY-Erythromycin, CAF-Chloramphenicol, SXT-Trimethoprim-sulfamethoxazole, CRO- Ceftriaxone, DA- Clindamycin, TE- Tetracycline, AMP-Ampicillin, Amox/cla- Amoxicillin/clavulanate, CAZ-Ceftazidime, CTX- Cefotaxime, MER- Meropenem*

**Magnitude of Extended spectrum beta lactamase**

Among the 67 Gram negative isolates, 52 (77.6%) were resistant to third generation cephalosporins (ceftriaxone, ceftazidime and cefotaxime). Based on the CLSI guideline (20), 43 isolates were checked for the confirmation of ESBL production by combined disk diffusion method. *Providencia* species, *Serratia* species, *Citrobacter*species and *E.cloacae* were excluded from ESBL testing. The overall prevalence of confirmed ESBL producing Gram negative isolates were 39/43 (90.6%). Among these ESBL producing isolates 25(64%) were found from neonates, 7(18%) from pediatrics and 7(18%) from adults blood culture. Most common isolates were *K.pneumoniae*16 (41.2%) followed by, *K.ozenae*10 (25.6%), *K.rhinose*8 (20.5%) and *E. coli*5(12.8%)(Figure 1).




***ESBL*-**Extended Spectrum Beta Lactamase.*

*Figure 1: Magnitude of Extended spectrum beta lactamase production in Gram negative isolates from blood culture of ICU patients at the University of Gondar Comprehensive Specialized Hospital from February to May 2018.*

**Risk factors associated with bacterial blood stream infections**

The association between blood stream infections (BSIs) and suspected risk factors were assessed using logistic regression. The result of multivariable analysis model in the binary logistic regression indicated that only age and sex were significantly associated with BSIs. Those neonates were 2.65 times more likely to develop bacterial BSI than adult age groups [AOR=2.65 (95% CI: 1.29, 5.42, p=0.008)]. Patients who are males were 2 times more likely to develop bacterial infection than females [AOR=2.0 (95% CI: 1.23, 3.27, p=0.005)] (Table 4).

## DISCUSSION

In the current study, the overall prevalence of bacterial isolates was 25% which is similar with other studies conducted in India (23% and 24.7%)(17, 18). But it is higher than another study in India (11.8%) (19), Pakistan (16%)(20), Jimma (15.8%)(21) and Gondar (18.2%) (22). This variation may be due to differences in target population, quality of the specimen, and techniques of bacteriology.

Gram negative bacterial isolates have been reported to be more than a fold (69.8%) to Gram positive isolates (30.2%).This result is consistent with a study conducted in Kanpur, India recorded that 60% of the isolates were Gram negative and the remaining 30% were Gram positive (17). However, our study differs from the North India study in which Gram negative and Gram positive bacteria were 39.3% and 53%, respectively (18). The difference may be attributed to the patients admitted to the ICU, as well as the volume of the specimen and methods of processing. A prevalence of 31.4% of bacteria isolates was recorded in neonate ICU, which is lower than a report

**Table4:** Risk factors associated with bacterial infection from blood culture of ICU admitted patients at the University of Gondar Comprehensive Specialized Hospital from February to May2018.

| Variables | | Organisms isolated | | | |
|---|---|---|---|---|---|
| | | Yes | No | COR (95% CI) | AOR (95% CI) |
| **Age** | Neonates | 65 | 142 | 2.47(1.22,5.00) * | 2.65(1.29,5.42)** |
| | Pediatrics | 20 | 93 | 1.13(0.5,2.54) | 1.10(0.50,2.50) |
| | Adult | 11 | 53 | 1 | 1 |
| **Sex** | Male | 62 | 143 | 1.84(1.1,2.9)* | 2.00(1.23,3.27)** |
| | Female | 34 | 145 | 1 | 1 |
| **Residence** | Rural | 73 | 194 | 1.54(0.9,2.6) | 1.57 (0.91,2.72) |
| | Urban | 23 | 94 | 1 | 1 |
| **Place of admission** | NICU | 65 | 145 | 2.48(1.20,5.00)* | 0.50 (0.01,21.28) |
| | PICU | 20 | 82 | 1.35(0.60,3.00) | 2.66 (0.15,46.76) |
| | AICU | 11 | 61 | 1 | 1 |
| **Admitted to ICU from** | Medical center | 85 | 228 | 2.03(1.01,4.00)* | 1.30 (0.59-,2.89) |
| | Home | 11 | 60 | 1 | 1 |
| **Prior antibiotic therapy** | Yes | 48 | 119 | 1.42 (0.8-2.2) | 2.61 (0.85-8.01) |
| | No | 48 | 169 | 1 | 1 |
| **Device used** | Yes | 46 | 126 | 1.18(0.7-1.8) | 0.43 (0.14-1.33) |
| | No | 50 | 162 | 1 | 1 |

***Keys:*=P-value <0.05, **=P-Value<0.01,****COR- Crude odds ratio, AOR-Adjusted odds ratio, NICU-neonatal intensive care unit, PICU- paediatric Intensive Care Unit, AICU-adult intensive care unit.*

in India (66.3%)(23) and Gondar, Ethiopia (46.6%) (24). *K.pneumoniae* (15.4%) was the most prevalent isolate in this ICU, close to a previous study in Gondar (15.8%) (24).This may be due to similar study area after a period of five years.

According to the present study, the predominant and the most common bacterial isolate of blood culture was *K. pneunoniae* which accounts for 18.8%. This finding is comparable to those studies conducted in India(19.7% and 22%) (17, 18) and higher than reported in Jimma (6.7%) (21).This variation may be due to the study population in Jimma was adults but in the current study most participants were neonates. The percentage of *E.coli* was 7.3% which is approximately similar with previous study done in Gondar (7%) (22) and Bangladesh (10.3%) (25) . However, the result of the present study is lower than a study conducted in India which is 22% (17).

Among Gram positive isolates coagulase negative *staphylococcus* (CoNS) were the most common isolates which accounts 13.5%,which is comparable with a report in Indonesia (14.9%)(4), Kanpur, India (15%) (17), and Jimma, Ethiopia (13.3%)(21) . These similarities may be due to CoNS are common sapro-

phyte which causes disease among immune-compromised patients. In this study majority of CoNS were isolated from neonate ICU which is a common cause of blood stream infection (BSI). *S.aureus* was second isolate which accounts 7.3%. This result was comparable with the study conducted in Bangladesh (5.3%) (25). However, it is lower than a study conducted in India (33.5%) (23) and Canada (21.1%) (26). The variation may be due to the different methodology used and the area of the study. The reason for this variation could also be the different types of samples such as wound discharge from ICU patients were used in a study conducted in Canada.

Among the seven *S.aureus* isolates, 57.1% were MRSA which was higher than other research works conducted in Jimma (33.3%) (21), India (48%) (23) and Saudi Arabia(43%)(27).This high resistance could be due to frequent use of beta-lactam dugs such as penicillin, inappropriate use of antibiotics and few people self prescribing antibiotics and treatment by the patients due to availability of antibiotics on the market in the study area. In the contrary, our result is lower than a study conducted in Pakistan (76%). The reason for this may be due to a difference in laboratory processing system in Pakistan that uses both manual and automated BACTEC system that may increase the detection rate (20).

In the current findings, the overall prevalence of extended spectrum beta lactamase (ESBL) producing bacteria was 90.6%. This result is higher than the finding in Mali (61.8%)(28), Addis Ababa, Ethiopia (78.57%)(29)and Jimma, Ethiopia (38.4%)(30).This may indicate that, there is inappropriate and/or incorrect administration of antimicrobial agents in empirical therapies and lack of appropriate infection-control strategies which can cause a shift to increase in prevalence of resistant organisms in the ICU settings. The prevalence of ESBL producing *K.pneumonia* was 41%, which is higher than a report in Mali (29.4%) (28). On the other hand, the prevalence of ESBL producing *E.coli* was12.8%, which is lower than a report in Mali (23.6%)(28), Jimma, Ethiopia (28.2%)(30)and in Dhaka, Bangladesh (81.8%) (31).

In this study, being neonate and male had significant association with the occurrence of bacterial BSI. Similar results was shown in United States as well as in Italy that shows a lower prevalence in women than in men (32, 33). A more prominent hormonal or an increased estradiol level may enhance immune function in females and may be responsible for a protective effect in female critically ill patients in terms of development of severe sepsis (33).

**Limitations**

Although this research contains major bacterial pathogens responsible for sepsis, due to lack of laboratory facilities, other pathogens such as anaerobic bacteria, fungus and viruses have not been included. This study was also limited to conducting carbapenemase production tests among Gram negative isolates, penicillin MIC method for *S.pneumonia* as well as being unable to distinguish *Neisseria* species due to lack of materials and/or reagents. Besides, this study may not reflect rates for all hospitals in Ethiopia.

## CONCLUSION

Gram-negative pathogens were the most common isolates in bloodstream infections. The magnitude and/or distribution of bacterial isolates was high in neonates followed by pediatrics and adult patients. *Klebsiella* species were the most prevalent bacterial

isolates in both neonates and pediatrics, but *E.coli* in adult patients. Clindamycin for Gram positive, ciprofloxacin and meropenem for Gram negative isolates were most effective. There was an alarmingly high production of extended spectrum beta lactamase in gram-negative isolates as well as detection of methicillin-resistance in *Staphylococcus aureus*. This result might be a reflection of inappropriate use of antibiotics, poor infection prevention control practice in the ICUs. The emergence of this antibiotic resistance may lead to prolonged hospital stay, unnecessary cost, and mortality. Factors such as age and sex were significantly associated with blood stream infection.

The current finding may be useful in formulating and/or updating treatment guidelines to avoid unnecessary use of antibiotics for patients with ICU and to inspire researchers to perform advanced studies on emerging antibiotic resistance challenges. The data on the changing antibiotic resistance trends is also important for disease control activities in ICU settings.

**Abbreviations**

**AICU**: Adult Intensive Care Unit; **BSI**: Blood Stream Infection; **CLSI**: Clinical Laboratory Standards Institute; **CoNS:** Coagulase negative *Staphylococcus*; **EONS**: Early Onset Neonatal Sepsis; **ESBL**: Extended Spectrum Beta Lactamase; **ICU**: Intensive Care Unit; **LONS**: Late Onset Neonatal Sepsis; **MRSA**: Methicillin-resistant *Staphylococcus aureus*; **NICU**: Neonatal Intensive Care Unit; **PICU**: Paediatric Intensive Care Unit.

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