Microbiological Profile of Neonatal Sepsis at a Maternity Hospital in Omdurman, Sudan

Manal Abdelaziz¹, Yassir Hamadalnil¹, Omaima Hashim², Tahane Bashir³, and E. S. Mahjoub⁴

¹Department of Microbiology, Maternity Hospital, Khartoum
²Department of Microbiology and Virology, National Center for Gastrointestinal & Liver Diseases, Khartoum
³Department of Microbiology, Radioisotope Center Khartoum, Sudan
⁴Department of Microbiology, Red Sea University, Port Sudan
⁵Department of Microbiology, University of Khartoum, Khartoum, Sudan

Abstract

Background: Neonatal sepsis is a clinical syndrome characterized by systemic signs of infection and accompanied by bacteremia in the first month of life. It is a major cause of morbidity and mortality in neonatal period. The study was conducted to determine microbiological profile and antibiogram of neonatal sepsis at Omdurman Maternity Hospital.

Methods: This was a cross-sectional hospital-based study involving 202 neonatal blood cultures at Omdurman Maternity Hospital during the period from April 2017 to April 2018. Specimens were cultured in Brain Heart Infusion broth followed by subculture of isolates on blood agar, MacConkey agar, and Chocolate agar and incubated aerobically at 37°C for 24 h. The isolates were tested for their susceptibility to antimicrobial agents using the Kirby Bauer disc diffusion method.

Results: Of 202 positive blood cultures, 130 cases (64.4%) were early onset and 72 cases (35.6%) were recorded for late onset sepsis. Gram-negative pathogens approaching (123, 60.9%). *Staphylococcus aureus* was the most common organism in both groups of neonatal sepsis being isolated from (71, 35.7%), followed by *Klebsiella pneumoniae* (43, 21.2%). Gram-negative organisms were sensitive to Imipenem (97.3%) and Meropenem (80.5%) and resistant to third-generation Cephalosporins (65.3%) and Amoxicillin/Clavulanic acid (91.4%). Gram-positive organisms were resistant to Cefotaxime (75%), Amoxicillin/Clavulanic acid (65.4%), and Clindamycin (68.2%); 91.6% of gram-positive isolates were sensitive to Vancomycin.

Conclusion: Gram-negative pathogens took the major spectrum of isolates. *Klebsiella pneumoniae* (21.2%) was the most frequent gram-negative organism. *Methicillin-resistant Staphylococcus aureus* (MRSA) (33.7%) was the most common isolate. Most of the isolates were multidrug resistant. The best choice for treatment is Vancomycin (8.4%) and Imipenem (2.7%) for gram-positive and gram-negative, respectively. Adherence to antibiotic policy, antimicrobial surveillance, and policy updating is necessary.
1. Introduction

Neonatal sepsis is considered one of the major causes of morbidity and mortality among term and preterm infants in neonatal intensive care units (NICU) [1]. Neonatal sepsis is a systemic infection occurring in infants less than 28 days of life and is classified into early onset and late onset neonatal sepsis. Early-onset neonatal sepsis (EOS) occurs within seven days of birth, while late-onset neonatal sepsis (LOS) occurs after the first seven days of life [2]. Neonatal infections are responsible for about 1.6 million deaths every year in developing countries [3]. Sepsis and meningitis are considered the most common causes of these deaths [4]. In developed countries, *Streptococcus agalactiae* (GBS) and *Escherichia coli* were the most common pathogens isolated in severe cases [5]. EOS is caused by GBS, *E. coli*, *Streptococcus viridans*, *Staphylococcus aureus*, and *Haemophilus influenzae* [6]. The infant may acquire the pathogen either *in utero* or intrapartum [7]. According to the World Health Organization (WHO), there were around four million neonatal deaths every year, about a quarter of them is due to neonatal sepsis/pneumonia alone [8]. In Sudan, the prevalence of neonatal sepsis is 17.5% and the mortality is 14.5% [9]. This study was conducted to determine the microbiological profile and antibiogram of neonatal sepsis at Omdurman Maternity Hospital.

2. Materials and Methods

The present study was a cross sectional, hospital-based study, conducted at Omdurman Maternity Hospital during the period April 2017–April 2018. In this period, a total of 202 positive blood samples were analyzed. Two ml of blood from neonate was dispensed into a sterilized universal bottle containing brain heart infusion broth to make a 1:10 dilution. It was collected from neonates under aseptic condition. Each sample was cultured in blood culture bottle and incubated aerobically at 37°C up to seven days. Regular observation of the bottle color, and turbidity, direct gram stain, and subculture were done to detect the positive ones. Regular subculture was done after one day of incubation, at 48 hours, and then at 96 hours. Each sample was subcultured on blood agar, MacConkey agar, and Chocolate blood agar and incubated aerobically at 37°C for 24 h. If there was no broth change or negative subculture for up to seven days, blood culture was considered as negative. Identification was done based on colonial morphology, gram stain, and standard biochemical tests. Antimicrobial susceptibility of isolated organisms was detected by using the Kirby Bauer disc diffusion method according to the Clinical and Laboratory Standard Institute (CLSI) [10]. Pure colonies
of isolates were emulsified in distilled water and inoculated on Muller Hinton agar. Antimicrobial discs were placed on the inoculated agar and incubated for 24 h at 37°C; then they were observed for zones of inhibition and determined as susceptible, intermediate, or resistant according to the CLSI [10]. The applied antibiotic for gram-positive cocci included oxacillin to check MRSA and for gram-negative rods *E. coli* ATCC 25922 was used as control strain. Data were analyzed by statistical package for social science SPSS software version 24. Chi-square tests were done. *P*-value < 0.05 was considered significant in comparative data.

### 3. Results

A total of 202 newborn babies were enrolled in this study. There were 130 cases (64.4%) of early-onset sepsis and 72 cases (35.6%) of late-onset sepsis, see Table 1. Among the positive blood cultures, gram-negative septicemias were 123(60.9%), while gram-positive were 75(37.1%). *Staphylococcus aureus* was isolated from 71(35.7%) in both groups of neonatal sepsis, followed by *Klebsiella pneumoniae* 43(21.2%) Table 2. Tables 3 and 4 show the result of antibiotics sensitivity tests.

| Bacteria                      | Frequency of Isolation | Early Onset | Late Onset |
|-------------------------------|------------------------|-------------|------------|
| *Streptococcus pyogenes*      | 1                      | 1           | 0          |
| *Klebsiella oxytoca*          | 1                      | 1           | 0          |
| *Candida albicans*            | 4                      | 0           | 4          |
| *Enterococcus faecalis*       | 2                      | 0           | 2          |
| *Proteus mirabilis*           | 1                      | 1           | 0          |
| *Salmonella spp.*             | 6                      | 3           | 3          |
| *S. epidermidis*              | 1                      | 0           | 1          |
| *Enterobacter spp.*           | 6                      | 5           | 1          |
| *Acinetobacter baumannii*     | 5                      | 2           | 3          |
| *Serratia marcescens*         | 7                      | 7           | 0          |
| *Citrobacter spp.*            | 4                      | 1           | 3          |
| *Pseudomonas aeruginosa*      | 18                     | 12          | 6          |
| MRSA                          | 68                     | 43          | 25         |
| *Staphylococcus aureus*       | 3                      | 2           | 1          |
| *Klebsiella ESBL*             | 11                     | 8           | 3          |
| *Klebsiella pneumoniae*       | 32                     | 19          | 13         |
| *E.coli ESBL*                 | 9                      | 7           | 2          |
| *E. coli*                     | 23                     | 18          | 5          |
| Total                         | 202                    | 130         | 72         |
TABLE 2: Frequencies of bacteria that cause neonatal sepsis in which MRSA is the most common organism (n = 202).

| Type of Bacteria          | Frequency | Percent |
|---------------------------|-----------|---------|
| **MRSA**                  | 68        | 33.7    |
| Klebsiella pneumoniae     | 32        | 15.8    |
| **E. coli**               | 23        | 11.4    |
| Pseudomonas aeruginosa    | 18        | 8.9     |
| **Klebsiella ESBL**       | 11        | 5.4     |
| **E. coli ESBL**          | 9         | 4.5     |
| Serratia marcescens       | 7         | 3.5     |
| Enterobacter spp.         | 6         | 3.0     |
| Salmonella spp.           | 6         | 3.0     |
| Acinetobacter baumannii   | 5         | 2.5     |
| Citrobacter spp.          | 4         | 2.0     |
| Candida albicans          | 4         | 2.0     |
| Staphylococcus aureus     | 3         | 1.5     |
| Enterococcus faecalis     | 2         | 1.0     |
| S. epidermidis             | 1         | 0.5     |
| Proteus mirabilis          | 1         | 0.5     |
| Klebsiella oxytoca        | 1         | 0.5     |
| Streptococcus pyogenes    | 1         | 0.5     |
| **Total**                 | 202       | 100.0   |

TABLE 3: Frequencies of antibiotic susceptibility of the gram-negative organisms.

| Antibiotics                | Sensitive | Resistant |
|----------------------------|-----------|-----------|
| Imipenem                   | 110 (97.3%)| 3 (2.7%)  |
| Meropenem                  | 95 (80.5%) | 23 (19.5%)|
| Cefotaxime                 | 43 (40%)  | 66 (60%)  |
| Amikacin                   | 45 (46.9%)| 51 (53.1%)|
| Ceftazidime                | 36 (34.7%)| 68 (65.3%)|
| Gentamicin                 | 32 (39.5%)| 49 (60.5%)|
| Amoxicillin/clavulanic acid| 6 (8.6%)  | 64 (91.4%)|
| Penicillin                 | 7 (13.5%) | 45 (86.5%)|

4. Discussion

Neonatal sepsis can be defined as a clinical syndrome characterized by systemic signs of infection and accompanied by bacteremia in the first month of life [11]. It is considered one of the major causes in neonatal morbidity and mortality [1]. The pathogens causing neonatal sepsis in developing countries differ from those in developed countries [12]. But overall the gram-negative organisms are more common [13].
| Antibiotics                        | Sensitive | Resistant |
|-----------------------------------|-----------|-----------|
| Penicillin                        | 6 (9%)    | 61 (91%)  |
| Vancomycin                        | 66 (91.6%)| 6 (8.4%)  |
| Amoxicillin/Clavulanic acid       | 12 (34.6%)| 40 (65.4%)|
| Ciprofloxacin                     | 26 (43.3%)| 34 (56.7%)|
| Cefotaxime                        | 2 (25%)   | 6 (75%)   |
| Clindamycin                       | 7 (31.8%) | 15 (68.2%)|
| Gentamicin                        | 45 (60.8%)| 29 (39.2%)|

In this study, gram-negative and gram-positive organisms encountered 60.9% and 37.1%, respectively, which is comparable to a study conducted in India [12], which reported that gram-negative and gram-positive organisms were 56% and 44%, respectively. Similar results were reported in Iraq [14] and Tanzania [15].

In this study, the EOS and LOS were 64.6% and 35.4%, respectively. This result is also comparable to a study conducted in India [16], which reported that EOS and LOS were 61.41% and 38.59%, respectively, further supported by another study done in India [17]. However, it differs from the study conducted in Tanzania [15] and Iraq [14]. *Staphylococcus aureus* was the most common isolate (35.1%) in both EOS and LOS in the current study, and 95% of *S. aureus* were *Methicillin-resistant Staphylococcus aureus* (MRSA). This result is similar to a study conducted by Zaidi et al. [18]. The second most common isolate was *Klebsiella pneumoniae* (21.2%), from which 25.58% were Extended Spectrum Beta Lactamase (ESBL). High resistance rate to cephalosporins can be contributed to the irrational use of these antibiotics. The presence of *Proteus mirabilis*, *Serratia marcescens*, *Klebsiella oxytoca*, and *Streptococcus pyogenes* isolates in the EOS and the absence of these isolates in the LOS was noted in this study.

Antimicrobial resistance now is worldly distributed and multidrug-resistant organisms are a global concern. Antibiotics susceptibility patterns differ from one country to another, so it is difficult to compare the susceptibility patterns between countries. In this study, gram-negative organisms showed a high sensitivity to Imepenem (97.3%) which is consistent to the local study conducted at Soba Hospital in Khartoum (100%) [19]. Imepenem sensitivity was 80.5%. This study reflected moderate resistance to Amikacin (53.1%) and Gentamicin (60.5%), which differs from the local study that showed low resistance to Amikacin (10.5%) and high resistance to Gentamicin (89.5%) [19]. This study reflected a moderate resistance to third generation cephalosporin 60% when it’s compared to the local study that reflected high resistance (89.5%) [19]. The best sensitivity of gram-positive organisms was to Vancomycin (91.6%), most of these isolates...
were MRSA. This result was comparable to Soba study (92%) [19]. Gentamicin showed moderate sensitivity (60%), which is also comparable to the Soba study (50%) [19]. Also, for the gram-positive organisms, the third generation cephalosporin showed a moderate resistance (75%). Most probably the overuse of third-generation cephalosporin is a main cause of the wide range resistance.

5. Conclusion

From this study, gram-negative pathogens took a major spectrum of isolates. *Klebsiella pneumoniae* was the most frequent gram-negative organism. *Methicillin-resistant Staphylococcus aureus* (MRSA) was the most common isolate. Most isolates were multidrug resistance. Therefore, the surveillance for antimicrobial resistance was necessary. Adhering to and updating the antibiotic policy contributes to containing this problem.

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