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Original Article

Isolation of Methicillin-resistant *Staphylococcus aureus* from Neonatal Sepsis at a Tertiary Care Hospital

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Abstract

Introduction: Septicemia is the significant cause of morbidity and mortality in the neonates and is responsible for 30-50% of total neonatal deaths .Each year in developing countries. It is estimated that 20% of all neonates develop sepsis and approximately 1% die of sepsis related causes.In India, according to National Perinatal Database the incidence of neonatal septicemia has been reported to be 30/1000 live births. The emergence of methicillin resistant Staphylococcus aureus (MRSA) in neonatal patients is increasing. Early diagnosis and appropriate therapy of septicemia is of utmost importance to prevent morbidity and mortality.

Aim and Study: It was to find out the bacteriological profile in neonatal sepsis and study their antimicrobial susceptibility pattern including detection of MRSA.

Methods: This study was conducted for a period of one year in the department of microbiology in a tertiary care hospital. A total of 283 blood samples were collected using sterile precautions They were processed following standard laboratory protocol. Antibiogram was done using appropriate antibiotics by Kirby-Bauer disc diffusion method. Isolated of Staphylococcus aureus were tested for methicillin resistance.

Results : Blood from 283 neonates with the clinical signs and symptoms of sepsis were collected and samples were processed. Out of which 96 (33.92%) were culture positive.Total 53 (55.2%) Staphylococcus aureus were isolated out of which 27 (50.94%) were MRSA (Methicliin Resistant Staphylococcus aureus).Acinetobacter spp.was isolated in 15 (15.62%) cases.Klebsiella spp. was isolated in 13 (13.54%) cases. Psedomonas spp. was isolated in 3(3.12%) case . Antibiotic sensitivity test of MRSA was done and all MRSA isolates were sensitive to Vancomycin.

Conclusion: Multidrug resistance among the isolates was common. Early diagnosis and institution of specific antibiotics after studying the sensitivity pattern will help in reducing neonatal morbidity and mortality and prevent emergence of drug resistant strains. An effective infection-control programme, regular antibiotic susceptibility surveillance ,evaluation, and the enforcement and periodic review of the antibiotic policy of the hospital as well as the encouragement of rational antibiotic use will reduce the rates of development of bacterial resistance.

Key words: Septicemia, Neonate, Methicillin, Vancomycin, Drug resistance

INTRODUCTION

In the developing world, septicemia in neonates is one of the major causes of morbidity and mortality among the newborns. It is defined as "a clinical syndrome which is characterized by systemic signs and symptoms and bacteremia during the 1st month of life." It is "early-onset" disease, if it presents during the first 5–7 days of life and it is "late onset" if it occurs after the 1st week of life.[1]

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Sepsis is the most common cause of neonatal mortality and each year in developing countries it is responsible for 30–50% of total neonatal deaths. It is estimated that 20% of all neonates develop sepsis and approximately 1% die of sepsis-related causes. National Neonatal-Perinatal Database

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from India has reported an incidence of the neonatal sepsis of 0.1–4.5% from 18 hospitals across India and the incidence of blood culture-proven sepsis was reported as 8.5/1000 live births.[2]

The factors which are associated with sepsis in newborns include low birth weight, fetal distress, a low Apgar score, the requirement of mechanical ventilation, umbilical catheterization, and a history of preeclampsia in the mothers.[1]

The microorganisms most commonly associated with early-onset septicemia include Group B *Streptococcus, Escherichia coli,* coagulase-negative *Staphylococcus* species (CONS), *Haemophilus influenza,* and *Listeria monocytogenes* and late-onset septicemia is caused by CONS, *Staphylococcus aureus, E. coli, Klebsiella spp., Pseudomonas spp., Enterobacter spp., Candida spp.,* GBS, *Serratia spp., Acinetobacter spp.,* and anaerobes. The recent trends show an increase in infections due to CONS. The knowledge of bacteriological profile and its antibiotic sensitivity patterns is of immense help in saving lives of neonates with septicemia.[4]

An early treatment and the appropriate and the rational use of antibiotics can minimize the risk of severe morbidity and mortality in neonatal sepsis, and reduce the emergence of multidrug-resistant organisms. For the success of an early empiric treatment, a periodic review of the cases to assess any changing trends in the infecting organisms and their antimicrobial susceptibility is important.

The emergence of methicillin-resistant *S. aureus* (MRSA) in neonatal patients is increasing. The aim of this study was to find out the bacteriological profile in neonatal sepsis and study their antimicrobial susceptibility pattern including detection of MRSA.

MATERIALS AND METHODS

This study was conducted for a period of 1 year in the department of microbiology in a tertiary care hospital.

Inclusion Criteria

Neonates clinically suspected of having sepsis, temperature >99°F or <95°F, respiratory rate>60/min, abnormal cry, refusal of feed, drowsy or unconscious, septic focus on skin or umbilicus, diarrhea, and seizures were included in the study [Chart 1].

Exclusion Criteria

Neonates already on antibiotics and with diagnosis of intrauterine infection and congenital anomalies were excluded from the study.

A total of 283 neonates (0–28 days) with the clinical signs and symptoms of sepsis were included in this study. The neonates with congenital malformations or dysmorphic features were excluded from the study. The neonatal septicemia was categorized according to its time of onset as early-onset sepsis (0–7 days) and late-onset sepsis (8–28 days). An informed consent was taken from the parents of the neonates before the performance of this study. All the blood cultures were collected from the peripheral veins by following proper aseptic precautions before any antibiotic therapy was started with. Blood specimens were collected aseptically into BACTEC blood culture bottles after cleaning proposed venipuncture sites with 70% alcohol, then povidone iodine, and finally, 70% alcohol to remove the iodine at the end of venipuncture. Approximately, 2–3 ml of blood was inoculated into BACTEC blood culture bottles. The inoculated bottles were transported immediately to the department of microbiology and they were incubated in BACTEC blood culture system. Gram stain and subcultures using MacConkey and blood agar plates were done from culture bottles where growths were indicated, other specimens were inoculated on MacConkey agar and blood agar and incubated at 35–37°C for 18–24 h. The colonies which were isolated were identified on the basis of their colony morphology their Gram staining patterns and their standard biochemical tests. The antibiotic sensitivity patterns of the isolates were studied using the Kirby–Bauer disc diffusion technique. Detection of MRSA isolates was done using 1 μg oxacillin disc on Mueller-Hinton agar supplemented with an additional 5% NaCl and cefoxitin disc (30 ug) diffusion test, and results were interpreted according to the Clinical and Laboratory Standards Institute guidelines.[5]

RESULTS

This study was conducted for a period of 1 year in the department of microbiology in a tertiary care hospital. Blood from 283 neonates was collected and samples were processed, of which 96 (33.92%) were culture positive.

A total of 53 (55.2%) *S. aureus* were isolated, of which 27 (50.94%) were MRSA. *Acinetobacter spp.* was isolated in 15 (15.62%) cases. *Klebsiella spp.* was isolated in 13 (13.54%) cases. *Pseudomonas spp.* was isolated in 3 (3.12%) cases. Other organisms were in 12 (12.5%) cases. Antibiotic sensitivity test of MRSA was done and all MRSA isolates were sensitive to vancomycin [Chart 2].

DISCUSSION

In the present study, blood from 283 neonates was collected and samples were processed, of which 96 (33.92%) were

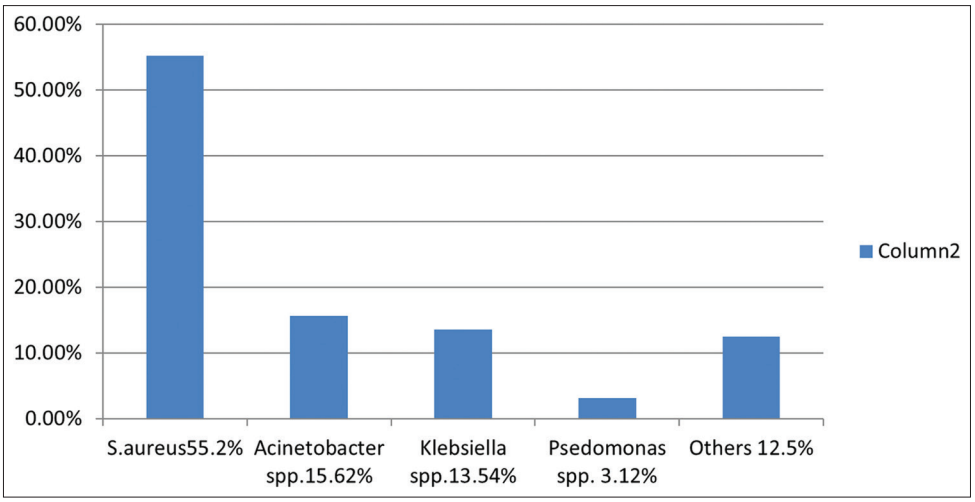

Chart 1: Organisms isolated from positive cultures of neonatal sepsis

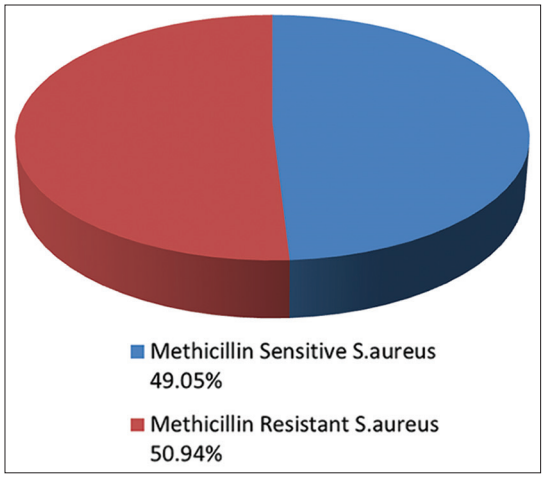

Chart 2: Methicillin sensitivity of *Staphylococcus aureus*

culture positive. A total of 53 (55.2%) *S. aureus* were isolated, of which 27 (50.94%) were MRSA

In the study done by Poonam Sharma *et al.*, a total of 131 organisms were isolated from the 311 blood cultures samples of neonates. Of these positive cultures, *S. aureus* were 68 (51.9%). Of these 68 *S. aureus* isolates, 39 (57.35%) were MRSA.[1]

In the study done by Chelliah *et al.*, of the 182 cases, 110 (60.4%) were culture positive. 30 (27%) were *S. aureus,* of which 17 (56.6%) of *S. aureus* were found to be MRSA.[2]

In a study done by Muley *et al.*, out of 180 blood samples, septicemia was confirmed by culture in 26.6% (48 of 180) of cases. Out of which *S. aureu*s was accounting for and 22.9% cases. 18.1% of *S. aureus* isolates were found to be methicillin resistant.[3]

In a study done by Thakur *et al.*, of a total of 450 neonates investigated with blood culture, 188 (42%) were found to be positive for neonatal septicemia. of 188 (42%) positive blood cultures, the Gram-positive bacteria and Gram-negative bacteria accounted for 60% and 40%, respectively. Among Gram-positive organisms, 66% isolates were *S. aureus,* of this methicillin resistance was detected in 29 (41%) of *S. aureus.*[4]

In a study done by Macharashvili *et al.*, 40% of *S. aureus* isolates were MRSA.[6]

In the study done by Ramesh Agrawal *et al.*, methicillin resistance prevailed in 61% (85/140) of coagulase-negative staphylococci and 38% (43/114) of *S. aureus* isolates.[7]

In a study done by Kung *et al.*, MRSA was detected in 12.8% of cases of the neonatal sepsis.[8]

In the study done by Pai, methicillin resistance was documented in 69 (29.1%) of 237 isolates.[10]

In the study done by Tiwari *et al.* among 783 isolates of *S. aureus*, 301 (38.44%) isolates were methicillin-resistant.[11]

Multidrug resistance among the isolates was common. Early diagnosis and institution of specific antibiotics after studying the sensitivity pattern will help in reducing neonatal morbidity and mortality and prevent emergence of drug-resistant strains.

In the present study, antibiotic sensitivity test of MRSA was done and all MRSA isolates were sensitive to vancomycin.

In the study done by Sharma *et al.*, most effective antibiotic against the MRSA isolates was vancomycin and sensitivity to vancomycin was 100%.[1]

In the study done by Chelliah *et al.*, 17 (56.6 %) of *S. aureus* were found to be MRSA and they were 100% sensitive to vancomycin.[2]

In a study done by Muley *et al.*, 18.1% of *S. aureus* isolates were found to be methicillin resistant. Vancomycin remains the drug of choice for MRSA strains.[3]

In a study done by Thakur *et al.*, methicillin resistance was detected in 29 (41%) of *S. aureus.* There were 30 (40%) MDR isolates among the total of 75 isolates of *S. aureus.* All the isolates were sensitive to vancomycin.[4]

In a study done by Nino *et al.*, 40% of *S. aureus* isolates were MRSA and all were susceptible to vancomycin.[6]

In the study done by Roy *et al.*, all *S. aureus* isolates were sensitive to vancomycin.[9]

In the study done by Tiwari *et al.* among 783 isolates of *S. aureus*, 301 (38.44%) isolates were methicillin-resistant.[11]

Therefore, regular surveillance of infections including antimicrobial susceptibility pattern of MRSA and formulation of a definite antibiotic policy may be helpful in reducing the burden of MRSA infections. Vancomycin should be used when the patient does not respond to the first-line treatment or the combination of drugs.

CONCLUSION

Neonatal sepsis is an important cause of neonatal mortality and it depends on the age of onset of sepsis and on the etiologic agent and their resistant pattern. Implementation of infection control measures, restricting the use of broad-spectrum antibiotics, rotation of antibiotics, and rationalizing the use of antibiotics can decrease antibiotic resistance. Early diagnosis and specific treatment can reduce neonatal mortality and morbidity. Therefore, regular surveillance of infections including antimicrobial susceptibility pattern of MRSA and formulation of a definite antibiotic policy may be helpful in reducing the burden of MRSA infections. Vancomycin should be used when the patient does not respond to the first-line treatment or the combination of drugs.

Multidrug resistance among the isolates was common. Early diagnosis and institution of specific antibiotics after studying the sensitivity pattern will help in reducing neonatal morbidity and mortality and prevent emergence of drug-resistant strains. An effective infection control programme, regular antibiotic susceptibility surveillance, evaluation, and the enforcement and periodic review of the antibiotic policy of the hospital as well as the encouragement of rational antibiotic use will reduce the rates of the development of bacterial resistance.

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