Microbial Profile of Neonatal septicaemia in a tertiary care hospital of Bhopal

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Abstract
Aims: To determine the microbial profile of neonatal septicaemia cases and antibiotic sensitivity pattern of aerobic isolates at a tertiary care hospital of Bhopal.

Material and methods: In this retrospective study the laboratory data of blood culture reports of 256 neonates admitted to NICU, People’s Hospital, Bhopal with clinical suspicion of septicaemia from July 2012 to December 2013 was analyzed.

Results: Total 256 samples were received during the study period of which 174 (68%) were found to be positive of which 118(67.8%), 50(28.7%) and 5(2.87%) had Gram positive, Gram negative and Candidal infection respectively. Staphylococcus aureus and Klebsiella pneumoniae were the commonest isolates. Gram positive organisms were found to be sensitive to Linezolid, Pristinomycin, Cefoxitin and Vancomycin. Imipenem, Levofloxacin and 3rd generation Cephalosporins were found to be effective against Enterobacteriaceae group. Pseudomonas and Acinetobacter sp. were found to be susceptible to Imipenem, Aztreonam and Piperacillin tazobactam.

Conclusion: This study emphasizes the need for longitudinal surveillance to be carried at regular intervals to describe the varied pathogens causing neonatal sepsis as well as their changing antibiotic susceptibility pattern which will provide a useful information base to guide practice and policies on rational use of antibiotics.

Keywords: Neonatal septicaemia, Blood culture, antibiotic susceptibility pattern.

1. Introduction
Neonatal sepsis is a clinical syndrome characterised by a constellation of non specific signs and symptoms in association with bacteremia during first month of life1.

It is an important cause of morbidity and mortality among neonates in India with an estimated incidence of approximately 4% in intramural live births2. Prompt recognition and appropriate antimicrobial therapy are the key determinants of positive outcome in this serious paediatric emergency1. Blood culture remains the gold standard for the diagnosis of neonatal septicaemia1,3,4,5.

A very wide spectrum of organisms has been described for cases of neonatal septicaemia and the spectrum is subjected to geographical alterations. Moreover the organisms isolated are often resistant to multiple antimicrobials which make treatment difficult. The uncertainty surrounding the clinical approach to treatment of neonatal septicaemia can be minimized by periodic epidemiological surveys of aetiological agents and their antibiotic sensitivity patterns. This will help clinicians provide safe and effective empirical therapies, develop rational prescription programmes and make policy decisions3,4,5,6.

With this background a retrospective study was undertaken to determine the aerobic microbial profile of neonatal septicaemia cases with antibiotic sensitivity pattern of isolates.

2. Materials and method
A retrospective study was conducted in the Department of Microbiology, People’s College of Medical Sciences and Research Centre, Bhopal wherein the blood culture data of 256 neonates admitted to Neonatal Intensive Care Unit, People’s Hospital with clinical suspicion of septicaemia from July 2012 to December 2013 was collected, reviewed and analysed to determine the microbial profile and antibiotic susceptibility pattern of aerobic isolates. This study was undertaken after seeking approval for conduct of study from Institutional Ethics Committee (Approval letter no.2986).

Blood specimens from the 256 neonates were drawn aseptically before starting any antibiotic therapy and inoculated in BacT Alert/PP. Plus blood culture bottles (Biomeurix India) which were incubated at 37°C aerobically in BacT alert 3D 120 automated blood culture system for 5 days. Growth if detected, were then subcultured on Mac Conkey’s agar and 5% Sheep blood agar by conventional method and was identified by standard microbiological techniques7,8.

Antimicrobial susceptibility testing was performed by Kirby Bauer Disc Diffusion method as per CLSI recommendations9.

2.1 Statistical analysis used
Data was maintained in Microsoft office Excel and tests of proportions were used for analysis.

3. Results
During the study period 256 blood samples from clinically suspected cases of neonatal septicaemia were obtained out of which Blood culture was positive in 174 cases, positivity rate being 68%.

The most common organism amongst Gram positive isolates was Staphylococcus aureus and amongst Gram negative isolates Klebsiella pneumoniae (Table 1).
The management of neonatal septicaemia cases, study of the bacteriological profile with their antibiotic susceptibility pattern plays a significant role. In this study blood culture positivity rate in neonatal septicaemia cases was 6%. Such high positivity rates were also reported by Ako Nai et al5 (55%), Shaw et al5 (54.6%), Tallur et al15 (64%) and Karthikeyan et al15 (51%).

Our study showed that Staphylococcus aureus was the most predominant isolate (54.6%). This observation is in accordance with the study conducted by Karki et al12 in Nepal, Karthikeyan et al15 in India, Meremikwu et al11 in Nigeria and Aziz et al15 in Pakistan.

In the present study we obtained 9.2% of coagulase negative staphylococci (CoNS) which was similar to study by Kumhar et al7.

Hammerberg et al16 had stated that if the venepuncture site had been carefully cleansed the growth of CONS in blood culture of specimens of premature neonates indicated bacteremia rather than skin contaminations in vast majority of cases. This view is also shared by Favre et al17 who concluded their study reporting that CoNS bacteremia harbour a significant mortality and a single positive blood culture in the presence of signs of sepsis should be considered as clinically relevant.

Table 1: Distribution pattern of microorganisms isolated from blood culture (n=174)

| Microorganisms                        | No. of isolates (%) |
|---------------------------------------|---------------------|
| **Gram positive Isolates**            |                     |
| Staphylococcus aureus                 | 119 (68.4)          |
| Coagulase negative Staphylococci      | 95 (54.6)           |
| Enterococcus                         | 16 (9.2)            |
| **Gram negative Isolates**            |                     |
| Escherichia coli                     | 6 (3.45)            |
| Klebsiella pneumoniae                 | 23 (13.22)          |
| Acinetobacter sp.                    | 9 (5.17)            |
| Citrobacter sp.                      | 4 (2.3)             |
| Pseudomonas aeruginosa                | 8 (4.6)             |
| Yeasts                                |                     |
| Candida sp.                           | 5 (2.87)            |

Note: Figures in bracket indicate percentages

The Antibiogram of the Gram positive and Gram negative isolates as per CLSI guidelines is depicted in Table 2 & 3

Table 2: Antibiotic sensitivity pattern of Gram positive isolates

| Drugs              | Staphylococcus aureus (n=95) | Coagulase Negative Staphylococci (n=16) | Enterococcus (n=8) |
|--------------------|------------------------------|-----------------------------------------|--------------------|
| Erythromycin       | 33 (35.3)                    | 3 (18.75)                               | 3 (37.5)           |
| Clindamycin        | 47 (49.5)                    | 13 (81.25)                              | 4 (50)             |
| Ciprofloxacin      | 68 (71.6)                    | 10 (62.5)                               | 3 (37.5)           |
| Ofloxacin          | 69 (72.3)                    | 11 (68.75)                              | 4 (50)             |
| Levofloxacin       | 70 (73.7)                    | 12 (75)                                 | 4 (50)             |
| Amikacin           | 36 (37.9)                    | 12 (75)                                 | 2 (25)             |
| Gentamycin         | 30 (31.6)                    | 11 (68.75)                              | NT                 |
| Cefotaxime         | 86 (90.5)                    | 13 (81.25)                              | 8 (100)            |
| Cefotaxime         | 35 (37)                      | 9 (56.25)                               | 3 (37.5)           |
| Cotrimoxazole      | 28 (29.5)                    | 6 (37.5)                                | 0                  |
| Ampicillin         | 12 (12.63)                   | 4 (25)                                  | 0                  |
| Linezolid          | 95 (100)                     | 16 (100)                                | 8 (100)            |
| Pristinomycin      | 95 (100)                     | 16 (100)                                | 8 (100)            |
| Vancomycin         | 95 (100)                     | 16 (100)                                | 7 (87.5)           |

Note: Figures in bracket indicate percentages. NT: Not Tested

Table 3: Antibiotic sensitivity pattern of Gram negative isolates

| Drugs              | K.pneumonia(23) | E.coli(6) | Citrobacter spp.(4) | Pseudomonas aeruginosa(8) | Acinetobacter spp.(9) |
|--------------------|----------------|-----------|---------------------|--------------------------|-----------------------|
| Amikacin           | 7 (30.43)      | 1 (4.17)  | 2 (50)              | 3 (37.5)                 | 4 (22.2)              |
| Gentamycin         | 5 (21.74)      | 1 (4.17)  | 2 (50)              | 2 (25)                   | 2 (22.2)              |
| Ampicillin         | 8 (35)         | 2 (33.33) | 2 (50)              | NT                       | NT                    |
| Amony-Clav         | 13 (56.52)     | 4 (66.67) | 2 (50)              | NT                       | NT                    |
| Ciprofloxacin      | 15 (65.2)      | 4 (66.67) | 2 (50)              | 3 (37.5)                 | 4 (44.44)             |
| Levofloxacin       | 16 (69.56)     | 6 (100)   | 4 (100)             | 4 (50)                   | 4 (44.44)             |
| Ceftriaxone        | 17 (74)        | 4 (66.67) | 2 (50)              | 4 (50)                   | 2 (22.22)             |
| Cefoperazone       | 2 (8.33)       | 2 (33.33) | 1 (25)              | 3 (37.5)                 | 3 (33.33)             |
| Cefazidime         | 19 (82.6)      | 5 (83.33) | 4 (100)             | 1 (25)                   | 2 (22.22)             |
| Cefotaxime         | 18 (78.26)     | 4 (66.67) | 3 (75)              | 1 (25)                   | 2 (22.22)             |
| Cefepime           | 4 (26)         | 4 (66.67) | 3 (75)              | 5 (62.5)                 | 4 (44.44)             |
| Imipenem           | 23 (100)       | 6 (100)   | 4 (100)             | 7 (87.5)                 | 7 (77.77)             |
| Cotrimoxazole      | 5 (21.7)       | 1 (4.17)  | 1 (25)              | NT                       | NT                    |
| Aztreonam          | NT             | NT        | NT                  | NT                       | NT                    |
| Piperacillin tazobactum | 16 (69.56)  | 4 (66.67) | 4 (100)             | 7 (87.5)                 | 7 (77.77)             |

Note: Figures in bracket indicate percentages. NT: Not Tested

Gram positive organisms were found to be most sensitive to Linezolid, Pristinomycin, Cefoxitin and Vancomycin. About 9.5% strains of Staphylococcus aureus were Methicillin resistant (MRSA).

Impenem, Levofloxacin and 3rd generation Cephalosporins were found to be most effective against Enterobacteriaceae group. Pseudomonas and Acinetobacter sp. were found to be most susceptible to Imipenem, Aztreonam and Piperacillin tazobactam

4. Discussion

For the effective management of neonatal septicemia cases, study of the bacteriological profile with their antibiotic susceptibility pattern plays a significant role.

In this study blood culture positivity rate in neonatal septicemia cases was 6%. Such high positivity rates were also reported by Ako Nai et al5 (55%), Shaw et al5 (54.6%), Tallur et al15 (64%) and Karthikeyan et al15 (51%).

Our study showed that Staphylococcus aureus was the most predominant isolate (54.6%). This observation is in accordance with the study conducted by Karki et al12 in Nepal, Karthikeyan et al15 in India, Meremikwu et al11 in Nigeria and Aziz et al15 in Pakistan.

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For *Staphylococcus aureus* 100% sensitivity was seen for Vancomycin, Linezolid, Pristinomycin that is in accordance with the study conducted by Mane et al.1. Vancomycin, Linezolid, Pristinomycin, Cefoxitin, Clindamycin and fluoroquinolones (Levofloxacin, ofloxacin, Ciprofloxacin) were highly effective against Gram positive organisms. Majority of Gram positive organisms were resistant to Amoxycillin.

About 9.5% strains of *Staphylococcus aureus* were Methicillin resistant (MRSA) which was similar to study by Kaitha et al.4. The result of antibiotic sensitivity pattern revealed that majority of Gram negative organisms were resistant to commonly used antibiotics.

Imipenem showed highest sensitivity for Gram negative organisms which was similar to study by Iregbu et al.5 and Rathod et al.6.

For Enterobacteriaceae group Imipenem showed 100% efficacy followed by third generation Cephalosporins (Ceftazidine-82.6%, Cefotaxime-78.26%, Ceftizoxime-74%) and Fluoroquinolones (Levoflox-69.56%, Ciproflox-65.2%).

Of the two aminoglycosides studied Amikacin scored over Gentamycin in terms of sensitivity for Gram positive as well as Gram negative organisms. This is in accordance to the study conducted by Kumhar et al.11 and Kairavi et al.5.

As this was a retrospective study of microbiological records it was not possible to correlate with neonatal morbidity and mortality and other markers of sepsis. The utility of this study would have definitely increased on inclusion of these data.

5. Conclusion

This study highlights the variable nature of antibiotic susceptibility patterns both in time and location around different geographical areas.

Amongst the cases of neonatal sepsis multiple antibiotic resistance is one of the greatest challenges creating hurdles in the effective management of infections. Slow speed in developing newer drugs and rapidity in resistance development are major areas of concern. Rational use of antibiotics should be done to avoid this.

Recommendations

This study stresses on the need for longitudinal surveillance at regular intervals to describe the pathogens causing neonatal sepsis as well as their changing antibiotic sensitivity pattern so as to rationalize a rational antibiotic policy.

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