

Original Research Article

A study of microbial pattern and antibiotic susceptibility in blood cultures among children aged upto thirty six months

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ABSTRACT

Background: Sepsis is the most common cause of high morbidity and mortality among newborn, infants and young children. The organisms implicated in these infections vary with geographical alteration. So antibiotics used should be decided by local prevalence of microbial pathogen, and its bacterial susceptibility pattern.

Methods: a prospective observational study among the different neonates and children aged 0 to 36 months. Blood samples are taken under aseptic precaution. Inoculated onto blood culture media (brain heart infusion broth). It is incubated at 37°C. Turbidity is observed daily and it is sub cultured on alternate days. If there is growth, the organism is identified by routine biochemical reactions and the antibiotic susceptibility test was done on the Muller Hinton agar using appropriate antibiotic discs. For testing antibiotic susceptibility, criteria defined by the clinical and laboratory standards institute (CLSI) were followed.

Results: Among 148 subjects 102 found to be culture positive with positive rate of 69%. Gram negative organism is more prevalence in this study, most common isolates from gram negative sepsis was *Klebsilla* species (22.2%) followed by *E. coli* (18.1%).

Conclusions: Gram negative organism form the majority of isolates in our setup with *Klebsilla* as the most common species among them, which was least sensitive to most of the drugs. Among gram positive organism MRSA as the most common isolates in our setup. Most of the antibiotic were sensitive. Limited and objective use of antibiotic therapy is a much-needed strategy and the new guidelines.

Key Words: Methicillin-resistant *Staphylococcus aureus*, Antibiotic susceptibility, Blood culture, Sepsis, Pediatrics, Clinical and laboratory standards institute

INTRODUCTION

Sepsis (symptomatic bacteremia) is the systemic maladaptive response to invasion of microbial organism in sterile body part, with a resultant high morbidity and mortality among pediatric population, because of their physiologically immature immune system. Maladaptive response means the circulating bacteria multiplying at a rate that exceeds their removal by phagocytes.^{1,2} Blood stream infections can affect any system of the body including respiratory system, gastrointestinal system,

central nervous system, cardiovascular system, genitourinary system, skin, and other systems. Septicemia can manifest in a wider range starting from self-limiting illness to sepsis, severe sepsis with organ dysfunctions, septic shock with cardiovascular dysfunction, life threatening conditions like consumption coagulopathy (DIC). Symptoms are produced by microbial toxins or cytokines produced by inflammatory cells.¹ In many studies' incidence were 20-50% positivity noted. It is a medical emergency that requires timely detection and urgent rational antimicrobial therapy.³⁻⁶ Clinical

presentation of pediatric sepsis varies for sites or system involved, so investigations are essential for early diagnosis and treatment. The organisms implicated in these infections vary with the geographical alteration. So, antibiotics empirically used should be decided by local prevalence of microbial pathogen, and its antimicrobial susceptibility pattern.⁷⁻⁹ The gold standard for diagnosis of septicemia is the isolation of bacterial agent from blood culture. Positive predictive value of blood culture is high enough to give an accurate diagnosis.¹⁰ Objective of the study is to determine the bacteriological profile of children with suspected septicemia and the antibiotic susceptibility pattern of the causative bacteria.

METHODS

A prospective observational study will be done in 148 children admitted in Government Cuddalore Medical College and Hospital, Chidambaram during the study period from January 2021 to March 2022 among the different neonates and children aged 0 to 36 months. Blood samples are collected under strict aseptic precautions. Around 1 ml of blood is drawn with sterile syringe and inoculated onto the blood culture media (Brain Heart Infusion Broth-10ml). It is incubated at 37°C. Turbidity is observed daily and it is sub-cultured on alternate days. If there is growth, the organism is identified by routine biochemical reactions and the antibiotic susceptibility test was done on the Muller Hinton agar using appropriate antibiotic discs. CRP is detected by latex agglutination test from serum sample of the subject as a sepsis marker. It will be correlated with clinical condition. Data on risk factors are collected by questionnaire. The Kirby-Bauer test, known as the disk-diffusion method, is the most widely used antibiotic susceptibility test in determining what choice of antibiotics should be used when treating an infection. This method relies on the inhibition of bacterial growth measured under standard conditions. For this test, a culture medium, specifically the Mueller-Hinton agar, is uniformly and aseptically inoculated with the test organism and then filter paper discs, which are impregnated with a specific concentration of a particular antibiotic, are placed on the medium. The organism will grow on the agar plate while the antibiotic “works” to inhibit the growth. If the organism is susceptible to a specific antibiotic, there will be no growth around the disc containing the antibiotic. Thus, a “zone of inhibition” can be observed and measured to determine the susceptibility to an antibiotic for that particular organism. The measurement is compared to the criteria set by the Clinical and Laboratory Standards Institute (CLSI). Based on the criteria, the organism can be classified as being Resistant (R), Intermediate (I) or Susceptible (S).

Inclusion criteria

Newborn and children aged 0 to 36 months with features suggestive of bacteremia admitted in department of pediatrics.

Exclusion criteria

Children and Newborns who are not having signs of bacteremia. Children and Newborn who have received parenteral antibiotics outside the hospital. Children more than 3 years of age. Parents/Guardians of the participants will be explained in prior about the study and informed consent will be obtained. Statistical analysis of data was done using statistical package for the social sciences (SPSS) 17 software.

RESULTS

Among the 148 subjects, 83 (56%) were male child and 65 (44%) were female child. Blood culture was done in our subjects. Among 54 (52.9%) males were culture positive and 48 (47.1%) female were culture positive. Methicillin resistant *Staphylococcus aureus* 45.45% was the most common organism isolated, which was mostly sensitive to Tigecycline 100%, Amikacin 86.6% and linezolid 62.2%. *Klebsilla pneumoniae* 22.1% was the second most common organism isolated, which was least sensitive to most of the antibiotics including Piptaz (50%), Meropenam (50%), Amikacin (54.5%).

Table 1: Gender distribution (n=148).

Gender	N	%
Male	83	56
Female	65	44

Between males and females in the overall blood culture growth positive rate (females 44% vs. males 56%, $p=0.2$) no significant difference among gender in this study.

Table 2: Anti-microbial susceptibility of *Klebsilla pneumoniae* (n=22).

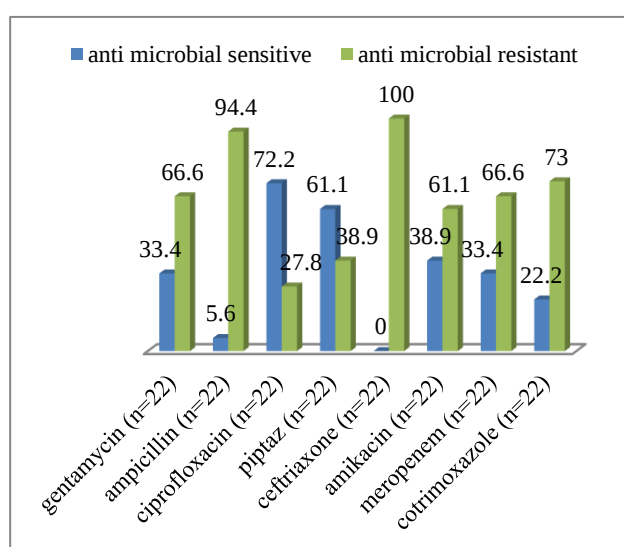
Antimicrobial	Anti-microbial susceptibility			
	Sensitive		Resistant	
	N	%	N	%
Gentamycin	9	40.9	13	59.1
Cotrimoxazole	3	13.6	19	86.4
Ciprofloxacin	10	45.4	12	54.6
Piptaz	11	50.0	11	50.0
Cefoperazone	6	27.3	16	72.7
Amikacin	12	54.5	10	45.5
Meropenem	11	50.0	11	50.0

DISCUSSION

In this study, between males and females in the overall blood culture growth positive rate (females 44% vs. males 56%, $p=0.2$) (Table 1, Figure 1). However, this finding disproves several other studies that reported high culture positivity in males compared with females. The reason for this difference is unclear and requires further study with large sample size. Bacteria are increasingly becoming resistant to conventional antibiotics in clinical and nonclinical settings.¹¹ The incidence of bacteremia varies widely in the pediatric population.

Table 3: Anti-microbial susceptibility of MRSA.

Antimicrobial	Anti-microbial susceptibility			
	Sensitive		Resistant	
	N	%	N	%
Gentamycin (N=45)	31	69.0	14	31.0
Erythromycin (N=45)	07	15.0	38	85.0
Clindamycin (N=45)	10	22.0	35	78.0
Cotrimoxazole (N=45)	15	33.0	30	67.0
Ciprofloxacin (N=45)	26	57.7	19	42.3
Linezolid (N=45)	28	62.2	17	37.8
Amikacin (N=45)	39	86.6	16	13.4
Cefoperazone (N=45)	21	46.6	24	53.4
Tigecycline (V=15)	15	100	0	0.0

**Figure 1: Antimicrobial susceptibility of *E. coli*.**

In our study, bacteremia's positivity came to be 69%, while a study in Nigeria showed the incidence of bacteremia to be 48.9% with *S. aureus* covering 48.7% of it.³ Our study reported the incidence of gram-negative bacteremia to be higher (54.6%) than gram-positive (45.4%). Gram negative sepsis was frequently noted in some studies like, Ali et al (63%), Sharma et al (72.7%).^{12,13} This difference can be explained by Varying degrees of medical practice, use of devices, variation from place to place. Most common isolates from gram negative septicemic children were *Klebsiella pneumoniae* (22.2%). These were the second predominant isolates found from overall culture positive isolates. Similar findings were observed in Nagussie et al, Dagnew et al.¹⁴ Overall antibiotic sensitivity for gram negative sepsis: majority of them were *Klebsiella* culture positive sensitive to Meropenem (50%), Piperacillin (50%), Amikacin (54.5%), gentamycin (40.9%), least sensitive to Cefoperazone (27.3%), Cotrimoxazole (13.6%) (Table 2, Figure 2). Among gram-positive isolates, Methicillin Resistant *Staphylococcus Aureus* (MRSA) with 45.45%,

similar to studies from Indian Network for Surveillance of Antimicrobial Resistance (INSAR) group, 41% MRSA, China & Gupta group 49.5%.^{14,15} Overall antibiotic sensitivity for gram positive sepsis: majority of them were sensitive to Tigecycline (100%), Amikacin (86.6%), Gentamycin (69%), Linezolid (62.2%), Cefoperazone (42.6%), Ciprofloxacin (57.7%) least sensitive to clindamycin (22%), erythromycin (15%) (Table 3, Figure 3). *Pseudomonas aeruginosa* is a motile gram-negative rod that belongs to the family *Pseudomonadaceae*, disseminated widely in nature, and it is highly prevalent in the hospital setting since it encourages bacterial growth.¹⁶ In our research, it exhibited 7.07% prevalence and marked resistance to several drugs. In our study, it showed sensitivity to piperacillin/tazobactam (71%), Meropenam (100%), ciprofloxacin (100%). The other gram-negative bacilli of our study included *E. coli* (18.1%), in our study it showed resistance to Ceftriaxone (100%), Ampicillin (94.4%), Meropenem (66.6%), these results are in agreement with other studies Bhatti et al (Table 4, Figure 4).^{11,17} Worldwide emergence of resistant bacteria is a real threat to a favorable outcome to conventional forms of infection in the hospital settings and community. A careful selection of antibiotics is required to avoid treatment failure and the spread of resistant strains. Poor infection control practices with inappropriate and unjust use of antibiotics are the main driving force behind the tremendous increase in antimicrobial resistance strategies like combination therapy, antibiotic restriction, and antibiotic cycling must be put in use to prevent the emergence of resistance. Association of public health policy with active research on the determinants of the evolution of resistance to antimicrobial agents and its reversal is much-needed action. This study should be done on a larger scale involving different departments of a tertiary care hospital to describe antibiotic resistance trends in all age groups.

CONCLUSION

Blood culture still remains the gold standard diagnostic test available for the treating physician to identify the pathogenic bacteria. The blood culture positivity rate in this study was 66.8%, with the prevalence being higher among neonates. The leading causes of septicemia were methicillin resistant *Staphylococcus aureus* and *Klebsiella* species. Blood cultures were found to be positive in 66.8% and negative in 33.2%. Methicillin resistant *Staphylococcus aureus* 45.45% was the most common organism isolated followed by *Klebsiella pneumoniae* 22.1%. Ciprofloxacin and Amikacin were found to be sensitive in most of the positive cultures. Ciprofloxacin and Meropenem were found to be 100% sensitive for pseudomonas positive cultures. Most of the drugs were found to be resistant to *Klebsiella* species positive cultures, Sensitive to Amikacin (55%), Meropenem (50%), Piperacillin and Tazobactem (50%), Ciprofloxacin (44%). Incidence of culture positive septicemia was equally in both males and females in our study. Early diagnosis will help the clinician to institute the

antibiotics promptly who will help in reducing the morbidity and mortality.

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