



BACTERIOLOGICAL PROFILE AND ANTIBIOTIC SUSCEPTIBILITY PATTERN OF GRAM NEGATIVE SEPTICEMIA IN A TERTIARY CARE HOSPITAL, KERALA- RETROSPECTIVE RECORD BASED STUDY

Microbiology

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ABSTRACT

Introduction - Sepsis the most serious infectious event and early identification of organism and its antibiotic sensitivity pattern will help the physician to treat the patient in sepsis. **Objectives**- To identify and analyse the antibiotic sensitivity pattern of gram negative organisms causing sepsis. **Materials and methods** - A retrospective record based study of two years was conducted in the Microbiology Department to identify the Gram negative organisms associated with sepsis and its antibiotic sensitivity pattern. **Results** - Of the 6286 blood samples cultured, 190 patients had Gram negative bacterial sepsis. The organisms isolated are *Escherichia coli* (37.4 %), *Klebsiella pneumoniae* (26.8 %), *Acinetobacter baumannii* (10 %), *Enterobacter cloacae* complex (8.4 %), *Burkholderia cepacia* complex (4.2 %), *Salmonella* spp. (4.2 %), *Burkholderia pseudomallei* (1.05 %), *Brucella melitensis* (0.5 %) and other nonfermenters (9.5 %). There is an increase in resistance of enterobacterales noted. **Conclusion**- *Escherichia coli* remains the major agent causing sepsis. Periodic evaluation of organisms and its antibiotic sensitivity pattern is essential to know the drug resistance pattern.

KEYWORDS

sepsis ,antibiotic sensitivity pattern ,*Escherichia coli*

INTRODUCTION

Sepsis is one of the most emergency and serious situation in clinical practice where early identification of organism its antibiotic sensitivity pattern from blood culture can save the patient. Some important factors in the detection of blood cultures include the number of samples, volume, collection site, use of automated systems for monitoring and type of incubation atmosphere (anaerobic or not). Other factors may interfere to give false positive or negative results, such as the use of antibiotics and laboratory limitations may delay diagnosis or give incorrect information¹.

Travancore Medical College Hospital is a tertiary care Centre with full fledged multispecialty departments and Microbiology laboratory attached to the Hospital has fully automated BacT /Alert blood culture and vitek 2 (Biomereaux) system of identification. So even the rare pathogens can be speciated and exact resistance pattern with MIC can be determined accurately. So this study is to assess the etiological agents of sepsis and to analyse the sensitivity and resistance pattern of isolates.

REVIEW OF LITERATURE

Sepsis is defined as the systemic inflammatory response to infection which is seen in 10 of 1000 hospitalized patients and multiple organ dysfunction syndrome (MODS) develops in 30% of these patients; mortality is observed in 20% of patients with sepsis and 60–80% of patients with septic shock. Early diagnosis and treatment are necessary due to high mortality rates².

World-wide, 13 million people develop sepsis each year, and as many as 4 million people have died. In 1996, there were 4.774 patients admitted to a teaching hospital in Surabaya, Indonesia, and 504 patients were diagnosed as having sepsis, with a mortality rate of 70.2%. A global effort is needed to improve the therapeutic management of sepsis because of its high prevalence and mortality rate³. Sepsis can result from bacteria, viruses, fungi, or parasites, or it can develop in noninfectious intraabdominal incidents such as severe trauma, pneumonia, pancreatitis etc.

Bacteria are the most common cause of sepsis. In different studies, it was reported that gram-negative bacteria were isolated 20–64% of the time in sepsis patients. The most frequent causes isolated in gram-negative bacterial sepsis are *E. coli*, *Enterobacter*, *Pseudomonas*, *Proteus*, *Acinetobacter*, *Klebsiella*, and other rare gram-negative bacteria, in order of their frequency⁴.

In various studies on neonatal sepsis, the common etiological agents are *Klebsiella pneumoniae*, *Enterobacter* spp., *Pseudomonas*

aeruginosa, *E. coli*, *Citrobacter* and *Salmonella Typhi*. It is very important to identify and analyse the antibiotic sensitivity pattern of blood stream isolates because the outcome of the patient is determined by the appropriate use of antibiotics⁶. Patients who received appropriate initial antimicrobial treatment have lower mortality than those of who didn't⁷. In other hand, wrong or inappropriate use of antibiotic will contribute to the development of antibiotic resistance and multi drug resistance (MDR). The high incidence of MDR can reduce the opportunities of patients to get the appropriate antimicrobial that can affect to increase the risk of death⁸. Raymond in his study have suggested a high mortality cases founded in the patients with MDR and the study also showed that the patients get inappropriate empirical antibiotic and severity of co-morbid⁹.

Aim

To identify and analyse the gram negative bacterial etiological agents of sepsis and its antibiotic susceptibility pattern in a tertiary care centre, Kerala

Objectives

1. To identify gram negative organisms causing sepsis from blood samples received in the laboratory for culture and sensitivity from 2017 July to 2019 June in Travancore Medical College, Kollam
2. To study the sensitivity and resistance pattern of gram negative isolates from blood culture samples

Materials and methods Study design-Retrospective record based study
Study setting -Department of Microbiology

Study population -Patients of all age groups whose blood samples were sent for culture

Study period -two years (2017 July to 2019 June)

Inclusion criteria- All blood samples received in the laboratory for culture and sensitivity during the time period 2017 July to 2019 June

Exclusion criteria-Samples with potential contamination; Organisms isolated from a single sample where primary cause is not known

Study procedure - After due clearance from the Institutional Ethics Committee, the WHONET software system, Medivare software and registers from 2017 July to 2019 June (two years) maintained in the Microbiology department were studied to identify the Gram negative organisms associated with sepsis and its antibiotic sensitivity pattern. The parameters included in the study are age, gender, department,

diagnosis, organism isolated and sensitivity pattern.

STATISTICAL ANALYSIS

The data was entered in Microsoft excel worksheet and the results were analysed by using simple descriptive statistics involving frequencies and proportions using SPSS¹⁶

ETHICAL CONSIDERATIONS

The study was conducted only after obtaining approval from the Institutional Ethics Committee. Confidentiality of the information obtained was assured throughout the study.

Funding: No funding sources

Conflict of interest: None

RESULTS

Of the 6286 blood samples cultured, 190 patients had Gram negative bacterial sepsis. 119 patients were males and 71 were females.

Table no 1-Distribution of organisms

Organism	Number	Percentage
Escherichia coli	71	37.4
Klebsiella pneumoniae	51	26.8
Enterobacter cloaca complex	16	8.4
Acinetobacter baumannii	19	10
Burkholderia cepacia complex	8	4.2
Salmonella Typhi/Paratyphi A(6+2)	8	4.2
Burkholderia pseudomallei	2	1.05
Brucella melitensis	1	0.5
*Other nonfermenters	18	9.5
Total	190	

*Other nonfermenters – Pseudomonas aeruginosa -4, Sphingomonas paucimobilis -5, Ochrobactrum anthropi-2, Stenotrophomonas maltophilia-5, Pasteurella maltocida -1, Pasteurella pneumotropica -1

Table no 2 -Distribution of patients according to location

Location	Number (%)
Emergency department	7(3.7%)
Ward	119(62.6%)
ICU	64(33.7%)

Table no 3-Sensitivity pattern of Enterobacterales-E.coli, Klebsiella and Enterobacter

Drug	E.coli % sensitive(n=71)	Klebsiella % sensitive(n=47)	Enterobacter
Ampicillin	6	0	0
Gentamicin	61	43	0
Cephalexin	11	13	0
Cefuroxime	11	21	19
Ceftriaxone	14	23	37
Cefepime	18	26	100
Ciprofloxacin	25	28	94
Trimethoprim/Sulfa methoxazole	44	30	94
Amikacin	76	36	88
Cefoperazone/Sulbactam	65	32	94
Piperacillin /Tazobactam	67	38	94
Imipenem	82	38	94
Meropenem	76	36	94

Table no 4 -Sensitivity pattern of Salmonella Typhi/Paratyphi A (n=6+2=8)

Antibiotic	Number (% sensitive)
Ampicillin	3(37.5%)
Ceftriaxone	4(50%)
Ciprofloxacin	4(50%)
Trimethoprim/Sulfamethoxazole	8(100%)

Azithromycin	8(100%)
Chloramphenicol	8(100%)

Table no 5-Sensitivity pattern of Acinetobacter baumannii (n=19)

Drug	% sensitive
Ampicillin	16
Gentamicin	47
cephalexin	16
cefuroxime	16
ceftazidime	16
Cefepime	21
Ciprofloxacin	68
Trimethoprim/Sulfamethoxazole	53
Amikacin	68
Cefoperazone/Sulbactam	63
Piperacillin /Tazobactam	63
Imipenem	68
Meropenem	63

Table no 6-Sensitivity pattern of B.cepacia (n=8)

Antibiotic	% sensitive
Ceftazidime	0
Levofloxacin	0
Trimethoprim/Sulfamethoxazole	50
Meropenem	100

Table no 7-Sensitivity pattern of Burkholderia pseudomallei(n=2)

Antibiotic	Number sensitive (%)
Ceftazidime	2(100%)
Piperacillin Tazobactam	2(100%)
Meropenem	2(100%)
Trimethoprim/Sulphamethoxazole	2(100%)
Levofloxacin	2(100%)

DISCUSSION

Of the 6286 blood samples cultured, 185 patients had Gram negative bacterial sepsis. 119 (63%) patients of total 190 were male patients and 71(37%) were females. Men had a higher number of blood culture positivity than women which is consistent with the study done by Kaur and Singh in 2014 who reported high culture positivity in 65.22% men. 10

The most common bacterial isolate is Escherichia coli (n=71) followed by Klebsiella pneumoniae (n=47). Escherichia coli remains the major cause of gram negative bacteremia in many studies. 11,12 62.7 % of patients were admitted in the ward followed by 33.7 % in the ICU at the time of sample collection. 3.7 % patients came to the Emergency Department as sepsis.

In the present study, E. coli, Klebsiella pneumoniae and Enterobacter spp showed 14 %, 23 % and 37 % sensitivity to Ceftriaxone. A study conducted by Saghir et al in 2009 showed that E.coli was 28% and Klebsiella was 22.44% sensitive to Ceftriaxone. 13

The study shows that sensitivity to amikacin for E. coli, Klebsiella spp., and Enterobacter spp. was 76%, 36%, and 88%, respectively. Ciprofloxacin sensitivity to E. coli, Klebsiella spp., and Enterobacter spp was 25%, 28%, 94% respectively. In a study conducted by Sammar Naveed et al in paediatric patients it was shown that Amikacin was 50 % and Ciprofloxacin is 35.1 % sensitive to gram negative isolates. 14

In this study E. coli, Klebsiella spp., and Enterobacter spp. showed 67%, 38%, and 94% sensitivity to piperacillin/tazobactam. Gram negative bacteria is 60.4 % sensitive to Piperacillin/tazobactam in a study conducted in paediatric patients. 14

Sensitivity of Carbapenems were found to be 83%, 38 % and 94% for E. coli, Klebsiella spp., and Enterobacter spp respectively. Carbapenem sensitivity to gram negative bacteria causing sepsis was 77.3 % for Imipenem and 67.5 % for Meropenem in a study conducted

in paediatric patients.14

The sensitivity of E.coli is in concordance with various studies. But there is an increase in resistance of , Klebsiella .Because the current study was conducted in a tertiary care centre where lots of patients are referred from other hospitals after the intake of antibiotics .Enterobacter spp is sensitive to most of the drugs.

There were 8 patients who were diagnosed with Enteric fever .Ceftriaxone and Ciprofloxacin were sensitive in 50 % cases whereas Azithromycin and Chloramphenicol were sensitive in all the isolates .In a study done by Kumar et al in 2007 ,it was concluded that Antibiotic sensitivity for cephalosporins was 96.2% while antibiotic resistance was 29.2% with fluoroquinolones. Sensitivity to chloramphenicol was 92.5%.But we cannot generalise the sensitivity pattern in the current study as the number of patients were very less. Decreasing susceptibility of Salmonella typhi to ciprofloxacin has been well documented in several studies and is indicative of the effects of indiscriminate use of this group of antibiotics.15,16

Acinetobacter baumannii complex is an increasingly identified pathogen causing blood stream infections especially in ICU patients . In this study 19 isolates were identified of which 16 % were sensitive to Ampicillin ,1st ,2nd and 3rd generation Cephalosporins. Cefepime was sensitive in 21 % of cases . Amikacin sensitivity was 68 % .Beta lactam/beta lactamase inhibitors were sensitive in 63 % isolates. Carbapenem was also sensitive in 63 % isolates . In a study conducted by Puri et al in 2014 , it was found that Carbapenem sensitivity is 57% followed by Amikacin -55.14 % .So the resistance is less in the current study compared to previous studies. 17

Burkholderia cepacia complex is an emerging pathogen for sepsis in Hospitals .In our study 8 patients had sepsis due to Burkholderia cepacia complex . The bacteria was 100 % sensitive to Meropenem and 50 % sensitive to Trimethoprim /Sulfamethoxazole. In India, there are no precise reports of the prevalence of BCC infection due to the lack of specific laboratory tests and, in most cases, these bacteria have been ambiguously reported as non fermenter or simply Pseudomonas spp. For this reason, reports of diseases due to these organisms are rare and it has been reported from few tertiary care centres in north India. Burkholderia cepacia complex has been recognised as the third most common nonfermenter over the last six years in PGIMER after P. aeruginosa and A. calcoaceticus-baumannii complex. 18,19

Brucellosis was diagnosed in a patient who had a history of travel and intake of raw goat milk from middle east. The organism was identified by Biochemical reactions and Vitek 2 identification system.

There were many other Gram negative organisms that were identified in Vitek 2. But the the current study excluded the contaminant organisms unless there was isolation from two samples or the patient had clinical suspicion of sepsis .

CONCLUSION

E.coli remains the major gram negative bacteria causing sepsis. There is an increase in resistance of Gram negative bacteria to antibiotics. Emerging pathogens like Burkholderia cepacia and Burkholderia pseudomallei are also increasing. So periodic evaluation of organisms and its antibiotic sensitivity pattern is essential to know the drug resistance pattern.

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