

Original Research Article

Study of Aerobic Bacteriological Profile and Antimicrobial Susceptibility Pattern among Isolates from Sterile Body Fluids

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ABSTRACT

Background: Infections of sterile body fluids are medical emergencies associated with high morbidity and mortality. Early identification of causative aerobic bacteria and their antimicrobial susceptibility patterns is crucial for timely, effective empirical therapy and for monitoring evolving antimicrobial resistance trends in tertiary care settings. The present study aims to evaluate the aerobic bacteriological profile and antimicrobial susceptibility pattern among isolates from sterile body fluids excluding blood.

Material and methods: Cross-sectional study was carried out for a period of 1 year. Clinical specimens (pleural fluid, peritoneal fluid, cerebrospinal fluid, synovial fluid, pericardial fluid) were subjected to cultural analysis according to standard laboratory procedures. Antibiotic sensitivity testing was done by Kirby Bauer Disc diffusion method as per CLSI Guidelines [M100, 30th edition, 2020].

Results: Culture positivity rate was 14.83%. Highest positivity rate was seen in pleural fluid (27.55%), followed by synovial fluid (23.07%), peritoneal fluid (17.85%), CSF (4.79%) and no bacterial growth among pericardial fluids. The most predominant isolates were *E. coli* (22%), *Pseudomonas* spp. (20%), *Klebsiella* spp. (15%), *Staphylococcus aureus* (13%), *Acinetobacter* spp. (9%) and *Enterococcus* spp. (5%). Enterobacteriaceae were sensitive to meropenem, amikacin and piperacillin/tazobactam. Gram-positive isolates were sensitive to vancomycin, linezolid. *Pseudomonas* spp. was sensitive to Polymyxin B, meropenem and piperacillin/tazobactam.

Conclusion: Timely and appropriate antibiotic treatment; often empiric, can improve the clinical outcome of body fluid infections. Hence, Regular monitoring of prevalent pathogenic organisms and their sensitivities are essential.

Keywords: Sterile body fluids, Bacteriological Profile, Antimicrobial susceptibility testing

INTRODUCTION

The human body is made up of 60% liquids, there are different types of body fluids serve to lubricate the parietal and visceral surfaces ensuring smooth functioning of the respective organ / organ system & play an important role in body functions by

transporting nutrients, in regulating the temperature of the body, in respiratory processes and in transporting waste products of the body.

Microorganisms like bacteria, fungi, virus and parasites may invade and infect the body fluids and result in severe morbidity and mortality.¹ Early detection and rapid identification of microorganisms

are crucial for the appropriate management. Availability of early information helps the clinician to initiate early and more specific treatment and reduced lengths of stay of the patients in the hospital with less adverse effects.²

For empirical treatment, awareness of local antimicrobial susceptibility pattern and causative bacteria is essential. There is a need of periodic analysis of the local geographical bacteriological profile and antibiotic susceptibility pattern of organisms isolated and the results need to be communicated to the clinician. So, the present study was undertaken to evaluate aerobic bacteriological profile along with their antibiogram from various sterile fluids in a tertiary care hospital.

MATERIAL AND METHODS

The cross-sectional study was carried out in the department of Microbiology, at tertiary care hospital. A total of 364 samples were analyzed. Pleural, peritoneal, cerebrospinal fluid (CSF), synovial & pericardial fluids were drawn using proper aseptic precautions and sent to Department of Microbiology and then data were further analyzed in pharmacology department.

INCLUSION CRITERIA: Sterile body fluids received for culture and sensitivity from different IPDs & OPDs irrespective of age and sex received in Microbiology Department.

EXCLUSION CRITERIA: All blood samples

Sample Processing: The samples collected were processed in our laboratory using Standard microbiological procedures.³ The samples were subjected for Gram Stain and Culture. The culture media used were Blood Agar, Chocolate Agar & Mac Conkey Agar to obtain isolated colonies. The isolated colonies were then identified by Gram Stain, Motility and Standard Biochemical tests.

Data were entered and analyzed using Microsoft Excel. Descriptive statistics were used to summarize the study findings, with categorical variables presented as frequencies and percentages. The bacteriological profile and antimicrobial susceptibility

patterns were expressed using appropriate tables and percentages.

Antimicrobial Susceptibility Test: The Antimicrobial sensitivity test was performed for isolated organisms by Kirby Bauer Disc Diffusion method according to CLSI guidelines [M100, 30th edition, 2020].⁴

Drugs for Gram Positive Cocci: Cefoxitin, Ciprofloxacin, Gentamicin, Tetracycline, Erythromycin, Clindamycin, Co trimoxazole, Linezolid and Levofloxacin, Vancomycin, Rifampicin, Chloramphenicol.

Drugs for Gram Negative Bacilli: Ampicillin, Ampicillin- sulbactam, Cefuroxime, Cefotaxime, Cefepime, Ceftazidime, Amikacin, Gentamicin, Meropenem, Levofloxacin, Ciprofloxacin

Drugs for Pseudomonas: Piperacillin-tazobactam, Ceftazidime, Cefepime, Amikacin, Gentamicin, Ciprofloxacin, Levofloxacin, Meropenem, Aztreonam, Polymyxin B.

RESULTS

A total of 364 body fluids samples were collected, majority of the general population from whom samples were obtained were male at (N=217) 59.61% in comparison to female who were at (N=147) 40.38%. Majority of samples were sent from inpatient department, from which, ICU (NICU, PICU, OBSTRETIC ICU, ICCU) unit seemed to dominate with 30%. Age distribution of culture positive cases revealed that 46.29% belonged to the age group 21-40 years.

Total of 364 body fluid samples were included in the study out of which CSF samples (167) constituted 46%, pleural fluid samples (98)27%, peritoneal fluid (56) 15%, synovial fluid (39)11% and pericardial fluid (4) 1% among them, 54 fluids showed growth with isolation rate of 14.83%, while others 310 (85.17%) showed no growth after 48 hours of incubation.

Table – 1: depicts the various types of body fluids processed along with their positivity rate.

Samples	Total no. Processed	No. Yielding growth	Percentage (%)
Cerebrospinal fluid	167	8	4.79%
Pleural fluid	98	27	27.55%
Peritoneal fluid	56	10	17.85%
Synovial fluid	39	9	23.07%
Pericardial fluid	4	0	0

Highest positivity rate was seen in pleural fluid, followed by synovial fluid, followed by peritoneal fluid, followed by CSF and no growth was seen in pericardial fluid.

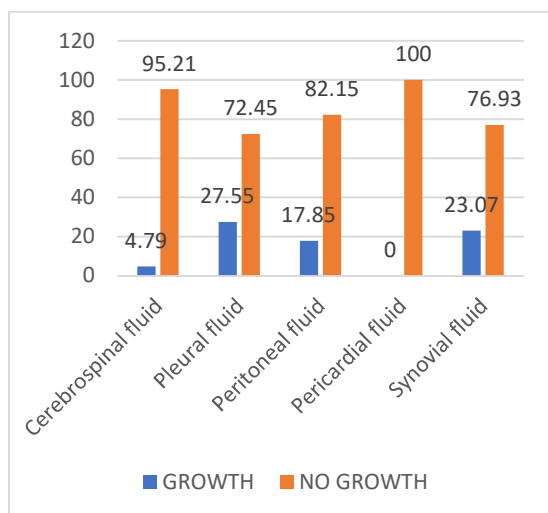


Figure – 1: Percentage distribution of various samples

Among total of 54 bacterial isolates, majority 40 (74.08%) were gram negative isolates, whereas 14 (25.92%) were gram positive isolates.

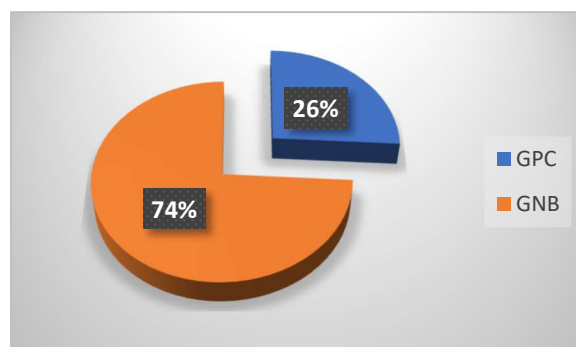


Figure - 2: Showing Distribution of Gram-positive and Gram-negative isolates

E coli was the most common bacterial organism overall, isolated from sterile body fluids with isolation rate of 22%, followed by pseudomonas aeruginosa (20%), followed by klebsiella pneumoniae (15%), staphylococcus aureus (13%), Acinetobacter spp. (9%), Enterococcus spp. (5%), followed by Proteus spp., CONS & Streptococcus spp. (4%), followed by Serratia spp. & Enterobacter spp. (2%).

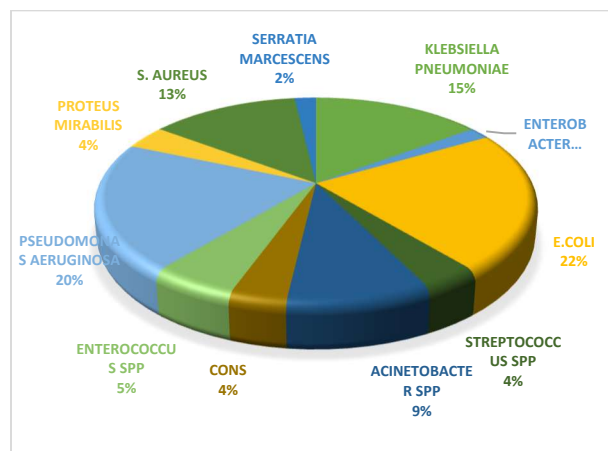


Figure - 3: Showing distribution of organism isolated

Table - 2: Bacterial profile in different body fluid samples

Organisms	Total Number	CSF	Pleural fluid	Peritoneal fluid	Synovial fluid
E. Coli	12	1	6	4	1
Klebsiella pneumoniae	8	2	3	2	1
Acinetobacter spp	5	3	2	0	0
S. Aureus	7	0	2	0	5
Streptococcus spp	2	1	0	0	1
Pseudomonas aeruginosa	11	0	8	2	1
CONS	2	1	1	0	0
Enterococcus spp	3	0	1	2	0
Enterobacter spp	1	0	1	0	0
Proteus mirabilis	2	0	2	0	0
Serratia marcescens	1	0	1	0	0
Total	54	8	27	10	9

Enterobacteriaceae were 100% sensitive to Meropenem (Fig-4), whereas Gram positive isolates were sensitive to Vancomycin, Linezolid (Fig-5). Non fermenter GNB showed good sensitivity to Meropenem & Piperacillin-tazobactam. (Fig- 6)

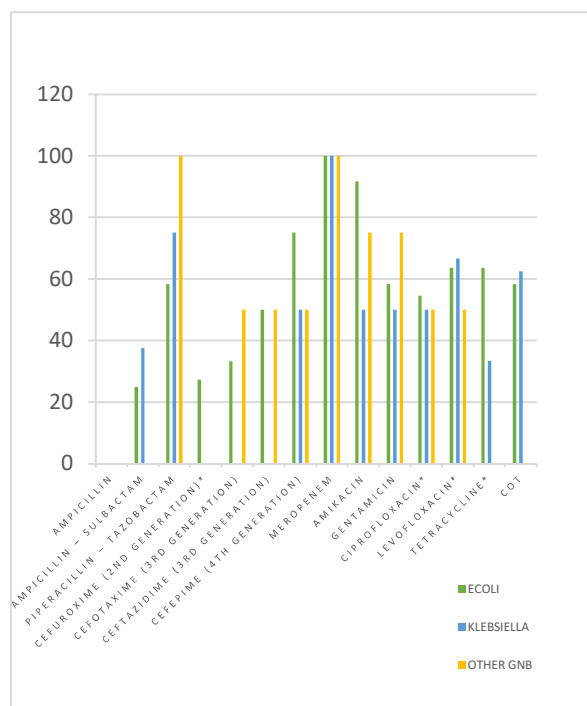


Figure – 4: Antibiotic sensitivity pattern among Enterobacteriaceae

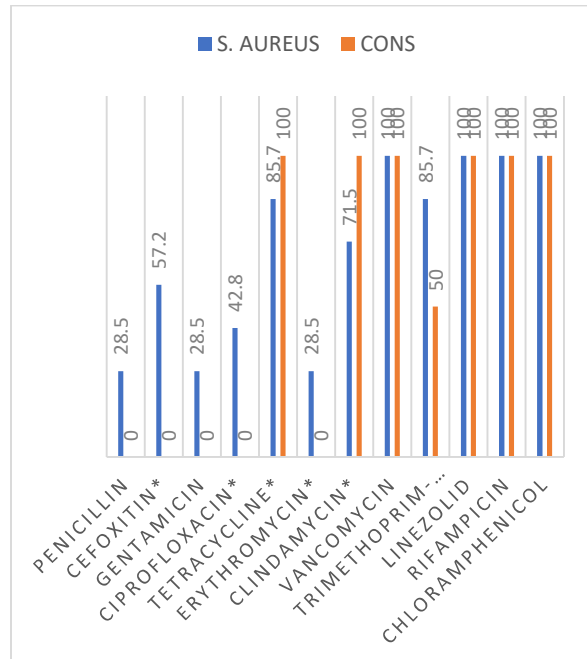


Figure - 5: Antibiotic sensitivity pattern of Gram-positive isolates

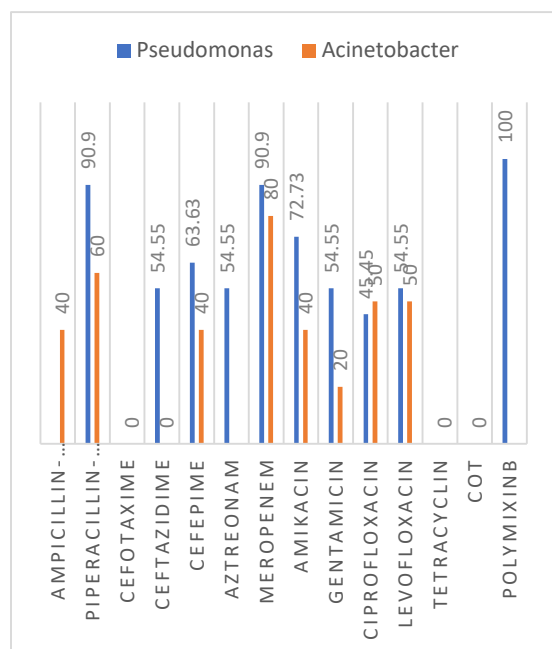


Figure – 6: Antibiotic sensitivity pattern of lactose non fermenter GNBS

*For cerebrospinal fluid, Fluoroquinolones, Tetracycline, 1st & 2nd generation cephalosporin, cephamycin & macrolides are not drug of choice and may not be effective treating CSF infections according to CLSI guidelines [M100, 30th edition, 2020].⁴ so excluded from denominator accordingly.

DISCUSSION

Sterile body fluids are those sites in which no microorganisms will usually grow, in a healthy or immunocompetent individual. However, if any of the organisms are isolated from these sterile fluids, then they are considered as significant and would be of life-threatening to the patient, either immunocompetent or immunocompromised.

Sterile body fluids include pleural fluid, peritoneal fluid, ascitic fluid, pericardial fluid, synovial fluid, cerebrospinal fluid. In present study, 54(14.83%) specimens were positive and 310(85.17%) were negative for the culture, isolation rate was 14.83%. A lower rate of positive cultures similar to the study has been observed in other studies like that of Deb A et al⁵ Rouf et al⁶ and Teklehymanot F et al⁷ which showed isolation rate of 14.41%, 10.81% and 14.1%

respectively. Positivity rate was quite low compared to other Indian studies such as Sharma et al⁸ & Sujatha et al⁹ which showed higher isolation rate of 30% and 31%. This might be due to over diagnosing, prior exposure to antibiotics and emergence of non-infectious conditions like malignancy, some of the variations are likely explained by the differences in the study population.¹⁰

It is observed that gram negative bacilli outnumber the gram-positive organisms. Similar predominance of gram-negative bacteria in sterile body fluids was also seen in the study of Rouf et al.⁶ Amongst gram negatives, *E. coli* was the most commonly isolated pathogen & amongst Gram positives, *Staphylococcus aureus* was most common, Rouf et al⁶ observed similar findings. In contrary to this, *Acinetobacter* spp. was the most commonly isolated pathogen in study done by Sharma et al.⁸

After the discovery and widespread use of antibiotics in the 1940s, *Staphylococcus aureus* succeeded *S. pneumoniae* and *S. pyogenes* as the major cause of empyema. Since the advent of beta-lactamase resistant semi-synthetic penicillins in the 1960s, the incidence of staphylococcal empyema has decreased and infections due to aerobic GNB as the predominant pathogen has increased markedly. Majority of positive cultures (66.66%) of peritoneal fluid were from adult patients from surgical wards. Bacterial peritonitis is a life-threatening infection in patients with cirrhosis and ascites and occurs in 10-30% of patients.¹² Most common bacterial isolate from synovial fluid was *S. aureus* (55.55%), Which is in agreement with study done by Nutt et al¹³. In our hospital very few pericardial fluid samples were received, only 1% compared to other body fluids & Pericardial fluids didn't yield any bacterial growth as bacterial infections of pericardium are relatively uncommon. Studies by Chandramukhi (1989)¹⁴, Chinchankar (2002)¹⁵, and Sonavane (2008)¹⁶ showed that the main organisms they found ie *N. meningitidis*, *S. pneumoniae* and *H. influenzae* in infants and children were not isolated in present study. This could be because of the increased uptake of childhood vaccines & increased awareness about pregnancy follow-ups & check-ups.

The reported spectrum of microorganisms responsible for body fluid infection is varied and is modified by introduction of antibiotics, patient specific factors such as surgical procedures, trauma or underlying conditions or by methodological factors namely the proper specimen collection, transport and culture. For these reasons, several studies have found discordant results in the spectrum of pathogens causing these infections.¹¹

E. coli isolates were sensitive to carbapenems (100%) & Amikacin (90%) & all isolates showed resistance to ampicillin, *A. baumannii* was least sensitive to all the antibiotics which was also seen in some other studies.⁶ In present study, resistance towards carbapenem among *klebsiella* spp. was not detected. Moderate resistance to aminoglycosides and beta-lactam beta-lactamase inhibitor combination seen. *Pseudomonas* isolates were highly sensitive to Meropenem (90.9%) and piperacillin-tazobactam (90.9%). It is in agreement with Sharma and Anuradha et al⁶. Among Gram positive isolates, *S. aureus* isolates were 100% sensitive to Vancomycin and Linezolid. No vancomycin resistant (VRSA) or Vancomycin-intermediate resistant *S. aureus* (VISA) isolates were detected in our study. No VRE (vancomycin-resistant enterococci) was detected in our study This may be due to judicious and controlled use of Vancomycin in the hospital.

Limitations: The study was conducted at a single tertiary care hospital, limiting generalizability. The sample size was relatively small, particularly for pericardial fluid. Only aerobic bacterial cultures were evaluated, excluding anaerobic, fungal and viral pathogens. Prior antibiotic exposure may have reduced culture positivity.

CONCLUSIONS

In the present study *E. coli* was the most common organism isolated from various body fluids. Significant numbers of both gram negative and gram-positive bacteria were isolated from various body fluid samples. Sensitivity of aerobic organisms to a particular agent varies from time to time and across different geographical area. Regular periodic evaluation of the bacteriological profile of body fluids

is essential, in any geographic location, along with antibiotic sensitivity pattern. This will help to empirically manage these life-threatening infections with the most appropriate antimicrobial that will be effective, thus reducing mortality and morbidity.

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