



Original Research Article

BACTERIOLOGICAL SPECTRUM AND ANTIBIOTIC RESISTANCE OF STERILE BODY FLUID INFECTIONS IN A TERTIARY CARE SETTINGR. Rajeswari¹

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ABSTRACT

Background: Infections involving sterile body fluids, such as cerebrospinal, pleural, peritoneal, synovial, and pericardial fluids, can lead to severe complications if untreated or improperly managed. Increasing antimicrobial resistance in these infections poses a critical challenge to patient care, especially in resource-limited settings like India. This study aims to assess the bacteriological profile and antibiotic susceptibility patterns of pathogens isolated from sterile body fluids in a tertiary care hospital, providing insights for empiric therapy optimization.

Material & Methods: This cross-sectional study was conducted on 661 sterile body fluid samples collected from patients with suspected infections at a tertiary care hospital in India from March 2022 to February 2024. Samples underwent Gram staining, culture on selective media, and standard biochemical tests for bacterial identification. Antibiotic susceptibility testing was performed using the Kirby-Bauer disk diffusion method, with interpretations based on Clinical and Laboratory Standards Institute (CLSI) guidelines. Data on patient demographics, clinical history, isolated pathogens, and resistance profiles were recorded and analyzed. Descriptive statistics were used to summarize prevalence rates, while chi-square tests were employed for comparing resistance rates, with significance set at $p < 0.05$.

Results: The most frequently isolated pathogens in our study were *Staphylococcus aureus* (13.4%), *Klebsiella pneumoniae* (14.5%), and *Escherichia coli* (13.0%), with notable resistance observed to commonly used antibiotics, including methicillin in *S. aureus* (45.5%) and third-generation cephalosporins in *K. pneumoniae* (58.3%) and *E. coli* (61.9%). Gram-positive bacteria showed high resistance to penicillin, while gram-negative isolates had a substantial prevalence of extended-spectrum beta-lactamase (ESBL) producers. The overall incidence of multidrug-resistant (MDR) organisms was high, emphasizing the need for targeted antibiotic stewardship strategies.

Conclusion: This study highlights the high prevalence of MDR pathogens in sterile body fluid infections, necessitating cautious empiric antibiotic selection. Findings underscore the importance of implementing routine antibiotic susceptibility testing and hospital-based antibiograms to enhance infection control practices and reduce treatment failures. Establishing effective antimicrobial stewardship programs is critical in limiting resistance trends and improving patient outcomes in resource-constrained healthcare settings.

Key Words: Sterile body fluids, antibiotic resistance, multidrug-resistant organisms, tertiary care hospital, *Staphylococcus aureus*, *Klebsiella pneumoniae*.

INTRODUCTION

Infections in sterile body fluids—such as cerebrospinal fluid (CSF), pleural, peritoneal, synovial, and pericardial fluids—are critical clinical concerns due to the severe health outcomes they can precipitate if untreated or inadequately managed.^[1] Such infections can lead to serious conditions, including meningitis, empyema, peritonitis, and septic arthritis, and are particularly challenging in intensive care units (ICUs), where they are reported to contribute to approximately 5-10% of cases.^[2] The mortality associated with these infections is also significant, with severe bacterial meningitis and sepsis resulting in death rates as high as 30% globally.^[3] These statistics underscore the need for rapid and accurate microbial identification and susceptibility testing to ensure prompt treatment, which is especially critical in resource-limited settings, where diagnostic limitations often hinder effective management.^[4]

The causative pathogens in sterile body fluid infections vary by region but frequently include *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Streptococcus pneumoniae*.^[5] In India, multiple studies report high resistance rates in these organisms, particularly to commonly used antibiotics.^[6] For instance, *E. coli* and *Klebsiella* spp. isolated from Indian hospitals often demonstrate resistance rates exceeding 60% to third-generation cephalosporins and fluoroquinolones.^[7] Additionally, *Staphylococcus aureus* shows significant methicillin resistance, with rates above 40% in some settings.^[8] This trend poses a considerable challenge to empiric therapy, as first-line treatments may not effectively target resistant strains, often resulting in extended hospital stays, higher healthcare costs, and poorer clinical outcomes.^[6,7]

Antimicrobial resistance (AMR) is a growing problem in India, compounded by widespread antibiotic misuse and over-prescription. Estimates suggest that up to 40% of antibiotic prescriptions may be unnecessary or inappropriate, further driving resistance.^[9] This issue is pronounced in tertiary care hospitals, where patients often present after multiple prior treatments, increasing the likelihood of multidrug-resistant (MDR) infections. Effective management of infections in these settings requires localized data on the prevalence of pathogens and their resistance patterns, given the geographic and institutional variability in microbial profiles and susceptibility trends.^[10]

This study aimed to characterize the bacteriological profile and antibiotic susceptibility patterns of infections in sterile body fluids at a tertiary care hospital in India. By identifying the specific pathogens and resistance trends in this population, the study will provide actionable insights that can guide empiric treatment and support more targeted

antibiotic use. These findings have the potential to inform local antimicrobial stewardship initiatives, reduce the inappropriate use of broad-spectrum antibiotics, and improve outcomes for patients with serious infections in sterile body sites.

MATERIALS AND METHODS

Study Design and Setting

This was a cross-sectional study conducted at Tertiary Care Hospital, located in North India. The study was carried out in the department of Microbiology over a period of 2 years between March 2022 to February 2024. This tertiary care hospital serves as a major referral center, catering to a diverse patient population from urban and rural settings, which provided a broad sample for assessing bacterial infections in sterile body fluids.

Study Population

Patients of all age groups and both sexes who presented with suspected bacterial infections in sterile body fluids including cerebrospinal fluid (CSF), pleural fluid, peritoneal fluid, synovial fluid, and pericardial fluid were eligible for inclusion. The inclusion criteria required that each sample collected was clinically indicated and obtained using standard aseptic techniques. Exclusion criteria were applied to any sample with evidence of contamination, insufficient quantity for analysis, or samples from patients who had received antibiotic therapy within 48 hours prior to sample collection.

Sample Collection and Processing

Sterile body fluid samples were collected following institutional aseptic protocols. CSF was obtained via lumbar puncture, while pleural, peritoneal, synovial, and pericardial fluids were aspirated under imaging guidance if needed, adhering to standard precautions. Each sample was immediately transported to the microbiology laboratory for processing.

Upon arrival at the laboratory, each sample was subjected to a series of diagnostic tests. Initially, direct Gram staining was performed to detect bacterial presence and assess the morphology of suspected pathogens. Following this, samples were cultured on blood agar, MacConkey agar, and chocolate agar plates, then incubated at 37°C, with observations for bacterial growth conducted at 24 and 48 hours. Finally, standard biochemical tests including catalase, coagulase, oxidase, and fermentation assays were performed to confirm bacterial identification based on both morphological and biochemical characteristics.

Antibiotic Susceptibility Testing

Isolated pathogens were subjected to antibiotic susceptibility testing using the Kirby-Bauer disk diffusion method according to Clinical and Laboratory Standards Institute (CLSI) guidelines.^[11] Antibiotic disks included commonly used agents such as cephalosporins (e.g., ceftriaxone), fluoroquinolones (e.g., ciprofloxacin),

aminoglycosides (e.g., gentamicin), and carbapenems (e.g., meropenem). Methicillin resistance in *Staphylococcus aureus* was evaluated using cefoxitin disks, and extended-spectrum beta-lactamase (ESBL) production was assessed in *Escherichia coli* and *Klebsiella pneumoniae* isolates.

Data Collection

For each patient included in the study, data were meticulously gathered on demographic variables (such as age, gender, and clinical history), sample type (CSF, pleural fluid, peritoneal fluid, synovial fluid, or pericardial fluid), and laboratory results in a preformed questionnaire.

Statistical Analysis

Data analysis was performed using statistical software SPSS version 25.0. Descriptive statistics, such as frequency distributions and percentages, were employed to summarize the prevalence of isolated pathogens and their respective antibiotic susceptibility patterns.

Ethical Considerations

The study protocol was reviewed and approved by the Institutional Ethics Committee. Written informed consent was obtained from all participants or their legal guardians. The study strictly adhered to ethical principles, maintaining patient confidentiality and ensuring that results were used solely for research purposes.

RESULTS

In this study of 661 cases involving sterile body fluids, the average age was 42.6 ± 16.7 years, with most participants aged 21-40 years (33.7%) and a male predominance (60.4%). Fluid samples included CSF (25.3%), pericardial fluid (26.6%), pleural fluid (19.4%), peritoneal fluid (16.2%), and synovial fluid (12.6%). Common underlying conditions were diabetes (26.3%), immunosuppression (19.1%), and CKD (15%). Clinical symptoms included fever (77.5%), pain (60.2%), respiratory (24.7%), abdominal (19.1%), and neurological symptoms (12.7%). The average hospital stay was 10.3 ± 4.6 days, with 18.3% in the ICU and a mortality rate of 11.8%. Bacterial growth was found in 24.8% of cases, with *Staphylococcus aureus* (13.4%), *Klebsiella pneumoniae* (14.6%), and *Escherichia coli* (12.8%) as common pathogens, highlighting the infection burden in patients with comorbidities and varied clinical presentations. [Table 1]

The distribution of pathogens across different sterile body fluids demonstrated variability. *Staphylococcus aureus* was isolated most frequently from synovial fluid (6.0%), followed by pleural fluid (3.9%), CSF (3.6%), and lower percentages in peritoneal (2.8%) and pericardial fluids (1.7%). *Escherichia coli* was most prevalent in peritoneal fluid (7.5%) and pleural fluid (4.7%), with lower frequencies in pericardial fluid (2.3%), synovial fluid (2.4%), and CSF (0.6%). *Klebsiella pneumoniae* showed higher prevalence in pleural

(5.5%) and peritoneal fluids (5.6%), with smaller proportions in other fluids. *Pseudomonas aeruginosa* was noted primarily in pericardial (2.8%) and synovial fluids (3.6%). *Streptococcus pneumoniae* appeared more frequently in CSF (4.2%) and pleural fluid (3.9%), while *Acinetobacter baumannii* was found in pleural fluid (2.3%) and pericardial fluid (1.7%). Lastly, *Enterococcus faecalis* was relatively more common in peritoneal fluid (3.7%) and had a consistent presence in pericardial, pleural, and synovial fluids at 2.3-2.4%. [Table 2]

The antimicrobial susceptibility patterns of pathogens revealed notable resistance across several antibiotics. *Staphylococcus aureus* exhibited the highest sensitivity to vancomycin (86.4%), followed by aminoglycosides (54.5%) and fluoroquinolones (45.5%). Resistance was highest to cephalosporins (50.0%). *Escherichia coli* demonstrated significant resistance to cephalosporins (61.9%), while showing high sensitivity to carbapenems (81.0%) and aminoglycosides (66.7%). Fluoroquinolones and aminoglycosides displayed intermediate resistance (14.3%-23.8%). *Klebsiella pneumoniae* also showed considerable resistance to cephalosporins (58.3%) and fluoroquinolones (50.0%), but was more susceptible to carbapenems (70.8%) and aminoglycosides (50.0%). *Pseudomonas aeruginosa* had significant resistance to cephalosporins (52.9%) and carbapenems (23.5%), with better sensitivity to piperacillin-tazobactam (64.7%) and aminoglycosides (52.9%). [Table 3]

The antibiotic susceptibility profiles of various pathogens showed a range of resistance and sensitivity patterns. *Staphylococcus aureus* was highly sensitive to vancomycin (86.4%), with moderate sensitivity to aminoglycosides (54.5%) and fluoroquinolones (45.5%), but exhibited significant resistance to cephalosporins (50.0%). *Escherichia coli* demonstrated high resistance to cephalosporins (61.9%) and moderate resistance to fluoroquinolones (33.3%) and aminoglycosides (23.8%), while being highly sensitive to carbapenems (81.0%). *Klebsiella pneumoniae* also showed considerable resistance to cephalosporins (58.3%) and fluoroquinolones (50.0%), with better sensitivity to carbapenems (70.8%) and aminoglycosides (50.0%). *Pseudomonas aeruginosa* exhibited significant resistance to cephalosporins (52.9%) and carbapenems (23.5%), but was more susceptible to piperacillin-tazobactam (64.7%) and aminoglycosides (52.9%). [Table 4]

The distribution of multi-drug resistant (MDR), extensively drug-resistant (XDR), and pan-drug-resistant (PDR) strains among the pathogens showed notable resistance profiles. Among *Staphylococcus aureus* (n=22), 45.5% were MDR, 18.2% XDR, and 4.5% PDR. *Escherichia coli* (n=21) exhibited 42.9% MDR, 9.5% XDR, and 4.8% PDR. *Klebsiella pneumoniae* (n=24) showed the highest MDR rate at 50.0%, with 20.8% XDR and 8.3% PDR. *Pseudomonas aeruginosa* (n=17) had 47.1% MDR, 23.5% XDR, and 5.9% PDR. *Streptococcus*

pneumoniae (n=16) had a lower MDR rate (31.3%) and only 6.3% XDR, with no PDR strains. *Acinetobacter baumannii* (n=10) had the highest

XDR proportion at 30.0%, with 60.0% MDR and 10.0% PDR. [Table 5]

Table 1: Demographic and Clinical Characteristics of Patients with Sterile Body Fluid Infections

Characteristic	Frequency (Mean ± SD)	%
Age (years)	42.6 ± 16.7	
Age Groups		
≤20 years	116	17.5
21-40 years	223	33.7
41-60 years	178	26.9
>60 years	144	21.8
Gender		
Male	399	60.4
Female	262	39.6
Type of Sterile Fluid		
CSF	167	25.3
Pleural Fluid	128	19.4
Peritoneal Fluid	107	16.2
Synovial Fluid	83	12.6
Pericardial Fluid	176	26.6
Underlying Conditions		
Diabetes	174	26.3
Immunosuppression	126	19.1
Chronic Kidney Disease (CKD)	99	15
Chronic Liver Disease (CLD)	46	7
Cancer	51	7.7
HIV/AIDS	29	4.4
Clinical Symptoms		
Fever	512	77.5
Pain	398	60.2
Respiratory symptoms	163	24.7
Abdominal symptoms	126	19.1
Neurological symptoms	84	12.7
Length of Hospital Stay (days)	10.3 ± 4.6	
ICU Admission	121	18.3
Mortality	78	11.8
Growth		
Yes	164	24.8
No	497	75.2
Pathogen		
<i>Staphylococcus aureus</i>	22	13.4
<i>Escherichia coli</i>	21	12.8
<i>Klebsiella pneumoniae</i>	24	14.6
<i>Pseudomonas aeruginosa</i>	17	10.4
<i>Streptococcus pneumoniae</i>	16	9.8
<i>Acinetobacter baumannii</i>	10	6.1
<i>Enterococcus faecalis</i>	14	8.5

Table 2: Distribution of Pathogens Isolated from Sterile Body Fluids

Pathogen	CSF (n=167)	Pleural Fluid (n=128)	Peritoneal Fluid (n=107)	Synovial Fluid (n=83)	Pericardial Fluid (n=176)
	Frequency (%)				
<i>Staphylococcus aureus</i> (n=22)	6 (3.6%)	5 (3.9%)	3 (2.8%)	5 (6.0%)	3 (1.7%)
<i>Escherichia coli</i> (n=21)	1 (0.6%)	6 (4.7%)	8 (7.5%)	2 (2.4%)	4 (2.3%)
<i>Klebsiella pneumoniae</i> (n=24)	3 (1.8%)	7 (5.5%)	6 (5.6%)	2 (2.4%)	6 (3.4%)
<i>Pseudomonas aeruginosa</i> (n=17)	2 (1.2%)	4 (3.1%)	3 (2.8%)	3 (3.6%)	5 (2.8%)
<i>Streptococcus pneumoniae</i> (n=16)	7 (4.2%)	5 (3.9%)	1 (0.9%)	2 (2.4%)	1 (0.6%)
<i>Acinetobacter baumannii</i> (n=10)	2 (1.2%)	3 (2.3%)	1 (0.9%)	1 (1.2%)	3 (1.7%)
<i>Enterococcus faecalis</i> (n=14)	1 (0.6%)	3 (2.3%)	4 (3.7%)	2 (2.4%)	4 (2.3%)

Table 3: Antibiotic Susceptibility Profile of Isolated Pathogens

Pathogen	Antibiotic	Sensitive	Intermediate	Resistant
		Frequency (%)		
<i>Staphylococcus aureus</i> (n=22)	Cephalosporins	8 (36.4%)	3 (13.6%)	11 (50.0%)

	Fluoroquinolones	10 (45.5%)	4 (18.2%)	8 (36.4%)
	Aminoglycosides	12 (54.5%)	2 (9.1%)	8 (36.4%)
	Vancomycin	19 (86.4%)	1 (4.5%)	2 (9.1%)
Escherichia coli (n=21)	Cephalosporins	6 (28.6%)	2 (9.5%)	13 (61.9%)
	Carbapenems	17 (81.0%)	1 (4.8%)	3 (14.3%)
	Fluoroquinolones	11 (52.4%)	3 (14.3%)	7 (33.3%)
	Aminoglycosides	14 (66.7%)	2 (9.5%)	5 (23.8%)
Klebsiella pneumoniae (n=24)	Cephalosporins	6 (25.0%)	4 (16.7%)	14 (58.3%)
	Fluoroquinolones	8 (33.3%)	4 (16.7%)	12 (50.0%)
	Aminoglycosides	12 (50.0%)	2 (8.3%)	10 (41.7%)
	Carbapenems	17 (70.8%)	2 (8.3%)	5 (20.8%)
Pseudomonas aeruginosa (n=17)	Cephalosporins	5 (29.4%)	3 (17.6%)	9 (52.9%)
	Piperacillin-tazobactam	11 (64.7%)	2 (11.8%)	4 (23.5%)
	Carbapenems	10 (58.8%)	3 (17.6%)	4 (23.5%)
	Aminoglycosides	9 (52.9%)	3 (17.6%)	4 (23.5%)

Table 4: Comparison of Antibiotic Resistance Patterns by Sample Type

Sample Type	CSF		Pleural Fluid		Peritoneal Fluid	
	Frequency (%)					
Pathogen	Staphylococcus aureus (n=22)	Escherichia coli (n=21)	Klebsiella pneumoniae (n=24)	Pseudomonas aeruginosa (n=17)	Acinetobacter baumannii (n=10)	Enterococcus faecalis (n=14)
Cephalosporins (%)	12 (54.5%)	9 (42.9%)	10 (41.7%)	8 (47.1%)	5 (50.0%)	N/A
Fluoroquinolones (%)	10 (45.5%)	7 (33.3%)	12 (50.0%)	10 (58.8%)	3 (30.0%)	3 (21.4%)
Aminoglycosides (%)	8 (36.4%)	6 (28.6%)	6 (25.0%)	9 (52.9%)	4 (40.0%)	4 (28.6%)
Carbapenems (%)	N/A	17 (81.0%)	17 (70.8%)	9 (52.9%)	6 (60.0%)	N/A
Vancomycin (%)	19 (86.4%)	N/A	N/A	N/A	N/A	11 (78.6%)
Piperacillin-tazobactam (%)	N/A	N/A	16 (66.7%)	9 (52.9%)	N/A	N/A

Table 5: Multidrug-Resistance Patterns in Pathogens Isolated from Sterile Body Fluids

Pathogen	MDR	XDR	PDR
	Frequency (%)		
Staphylococcus aureus (n=22)	10 (45.5%)	4 (18.2%)	1 (4.5%)
Escherichia coli (n=21)	9 (42.9%)	2 (9.5%)	1 (4.8%)
Klebsiella pneumoniae (n=24)	12 (50.0%)	5 (20.8%)	2 (8.3%)
Pseudomonas aeruginosa (n=17)	8 (47.1%)	4 (23.5%)	1 (5.9%)
Streptococcus pneumoniae (n=16)	5 (31.3%)	1 (6.3%)	0 (0.0%)
Acinetobacter baumannii (n=10)	6 (60.0%)	3 (30.0%)	1 (10.0%)

DISCUSSION

The present study, examining the bacteriological profile and antibiotic susceptibility patterns of pathogens in sterile body fluids, reveals crucial insights into the prevalence of multidrug-resistant (MDR) organisms and the challenges they pose in clinical management. This issue is especially pressing in the Indian context, where resource constraints and high antimicrobial use rates contribute to elevated resistance levels.^[12]

Our study found that *Staphylococcus aureus* was the most commonly isolated pathogen (13.4%), aligning with prior reports in Indian tertiary care centers, which also identified *S. aureus* as a leading pathogen in CSF, synovial, and pleural fluid infections. For instance, Patil et al., documented a similar prevalence of *S. aureus* in sterile fluids, with nearly 50% of isolates showing methicillin resistance (MRSA).^[13] Notably, *S. aureus* showed a high rate of multidrug resistance (45.5%) and a significant proportion of methicillin-resistant strains

(18.2%), consistent with findings from Khara et al., who highlighted MRSA as a significant concern in Indian hospitals due to limited therapeutic options.^[14] This high prevalence of MRSA underscores the necessity for vigilant infection control measures and targeted antibiotic strategies to mitigate MRSA transmission in hospital settings.^[12] Gram-negative bacteria, particularly *Klebsiella pneumoniae* and *Escherichia coli*, were also prominent in our study, with resistance rates above 50% to third-generation cephalosporins. The resistance rates are in agreement with studies conducted in similar settings across Southeast Asia and Africa.^[15] In our study, both organisms showed high MDR rates (50% for *K. pneumoniae* and 42.9% for *E. coli*), with *K. pneumoniae* also displaying significant resistance to fluoroquinolones (50%) and carbapenems (20.8%). In a study by Kar et al., 65% of *K. pneumoniae* and 58% of *E. coli* isolates were ESBL-producing, and both showed substantial resistance to cephalosporins and fluoroquinolones.^[16] Our findings echo these

observations, with both organisms exhibiting resistance due to the widespread use of beta-lactam antibiotics in empirical therapy, a point also raised by a study Sumbana et al., observed rising resistance levels associated with unchecked antibiotic distribution.^[17]

The presence of *Pseudomonas aeruginosa* in 10.4% of cases, with resistance rates to fluoroquinolones (52.9%) and carbapenems (23.5%), reflects the pathogen's inherent resistance mechanisms. A review by Sathe et al., found fluoroquinolone resistance in *P. aeruginosa* ranging from 50% to 70% across South Asian hospitals, with carbapenem resistance showing similar trends.^[18] This pathogen's adaptive resistance mechanisms, including efflux pump activation and beta-lactamase production, contribute to treatment challenges, as also demonstrated in study by Elfadadny et al., who emphasized the role of biofilm formation in conferring resistance.^[19]

Interestingly, *Streptococcus pneumoniae* was less frequently isolated in our study (6.3% of cases). This could indicate regional variation in pneumococcal carriage or reflect effective vaccination efforts, which have been shown to reduce invasive pneumococcal disease (IPD) prevalence.^[20] A study by Grant et al., noted a decrease in IPD among vaccinated populations, with a lower incidence of resistant strains.^[21] However, Sharma et al., reported persistent resistance in *S. pneumoniae* isolates in the North Indian region, especially to penicillin, necessitating ongoing surveillance of resistance patterns to adapt local empiric therapies accordingly.^[22]

The overall high prevalence of MDR pathogens in sterile fluids observed in our study aligns with the antimicrobial resistance trends reported globally, which emphasize the role of antibiotic stewardship programs. According to the Antimicrobial Resistance Collaborators., nearly 30% of infections in ICUs worldwide are due to MDR organisms, with the highest burdens in low- and middle-income countries, including India (WHO, 2020).^[23] Such trends complicate clinical management, as clinicians are increasingly forced to rely on reserve antibiotics, often associated with adverse effects and higher costs.^[24]

The clinical implications of our findings are significant: the presence of high resistance levels in pathogens isolated from sterile fluids necessitates a strategic revision of empiric treatment protocols.^[25] Standardizing antibiotic susceptibility testing and implementing localized antibiograms can support more effective initial therapy, minimizing broad-spectrum antibiotic use and improving patient outcomes. Additionally, our findings reinforce the need for robust infection control measures and the rational use of antibiotics, particularly in high-burden healthcare facilities.^[26]

CONCLUSION

In conclusion, this study provides a comprehensive overview of the bacteriological profile and resistance patterns in sterile body fluid infections in a tertiary care setting. The alignment of our results with both regional and global data highlights the critical need for localized, data-driven approaches to managing these infections, with a focus on enhancing antibiotic stewardship to curb the rise of multidrug-resistant pathogens.

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