ORIGINAL ARTICLE e-ISSN: 2349-0659 p-ISSN; 2350-0964

A Culture-antibiogram of Sterile Body Fluids in a Tertiary Care Hospital at Central Delhi in Emergency Settings

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Abstract

Infection of sterile body fluids could be life-threatening. Monitoring of prevailing infectious agents and their resistance pattern through antibiogram helps in appropriate selection of antibiotics and strengthens antibiotic stewardship. The aim of this study is to isolate infective bacteria from sterile body fluids and to determine antibiotic resistant pattern of these isolates. This retrospective observational study was conducted through January 2018–December 2018 in a tertiary care hospital at central Delhi. Sterile body fluids were processed as per the standard laboratory procedures. Antibiotic susceptibility pattern was determined by Kirby Bauer's disk diffusion method and interpreted as per Clinical and Laboratory Standards Institute guidelines. A total of 3,703 fluid samples were studied. Out of them 49.9% were ascitic fluid, 39.4% were pleural fluid, and 10.7% were others such as pericardial fluid, synovial fluid, bile, peritoneal dialysis fluid, and bronchoalveolar lavage fluid. Bacterial isolation rate was 12.93%. Predominant organisms were *Escherichia coli* (31%), *Acinetobacter* spp. (20%), *Klebsiella* spp. (16%), *Staphylococcus aureus* (11%), and *Enterococcus spp*. (10%). Isolation of vancomycin resistant enterococcus was 9.5% and Methicillin resistant *S. aureus* was 15.6%. Gram-negative isolates were 100% sensitive to colistin, followed by imipenem and gentamicin. Increased resistance shown toward third generation cephalosporins, amoxicillin + clavulinic acid, and piperacillin + tazobactum. *Klebsiella* spp. was the most resistant isolate, while *Enterococcus* spp. was the most resistant among gram positive isolates. Changing antimicrobial pattern poses challenge in treating infective agents. This culture antibiogram helps in appropriate selection of antibiotics in our setting and eventually decrease antibiotic resistant and patient's morbidity and mortality.

Keywords: Antibiotic stewardship, Antibiotic susceptibility, *Escherichia coli*, Fluid-antibiogram, *Staphylococcus aureus*

Asian Pac. J. Health Sci., (2022); DOI: 10.21276/apjhs.2022.9.4.21

Introduction

Body fluids are usually sterile. Some common pathogenic bacteria may invade the sterile body fluid to cause significant morbidity and life-threatening infections.[1] Sterile body fluid infections are medical emergency and need an early diagnosis and effective treatment.

The hospital antibiogram is a periodic summary of antimicrobial susceptibilities of local bacterial isolates. Antibiograms are often used by clinicians to assess local susceptibility rates as an aid in selecting an empiric antibiotic therapy and in monitoring resistance trends over time within an institution.[2] Periodic surveillance and knowledge of prevailing strains and their antibiotic susceptibility are helpful for framing the antibiotic policy and better management of patients.

The paucity of studies on antibiotic susceptibility and bacteriological profile of body fluids particularly in patient presenting with emergency leads to indiscriminate use of unnecessary antibiotics and eventually more antibiotic resistance. Hence, a culture antibiogram of body fluids is very crucial to clinicians, Microbiologists, Pharmacists, and Policy makers for proper diagnosis of different infections and for prudent antibiotic use.[3]

Thus, the aim of this study is to evaluate bacteriological profile and antibiotic susceptibility pattern of various body fluid collected from patients in a tertiary care hospital.

Materials and Methods

The present retrospective observational study was conducted through January 2018–December 2018 in emergency microbiology laboratory in a tertiary care hospital at central Delhi associated with a medical college.

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How to cite this article: Suman U, Lal R, Chaudhary M, Baveja CP. A Culture-antibiogram of Sterile Body Fluids in a Tertiary Care Hospital at Central Delhi in Emergency Settings. Asian Pac. J. Health Sci., 2022;9(4):99-103.

Source of support: Nil

Conflicts of interest: None.

Received: 12/02/2022 **Revised:** 10/03/2022 **Accepted:** 13/04/2022

A total 3,703 various body fluids were analyzed. Different body fluids such as ascitic fluid, pleural fluid, pericardial fluid, synovial fluid, bile, peritoneal dialysis fluid, and bronchoalveolar lavage fluid except Cerebrospinal fluid (CSF) were collected under proper aseptic precaution and processed within 2 h.

Inclusion Criteria

Sterile body fluids received from intensive care units (ICU), emergency ward, other in patient department (IPD) and outpatient department were included irrespective of age and sex.

Exclusion Criteria

The following criteria were excluded in the study:

1. Blood
2. CSF
3. Contaminated samples
4. Samples received after 2 h of collection

Sample Processing

Samples collected were processed in emergency laboratory using standard microbiological procedures. All the samples were subjected for Gram stain and aerobic culture at 37°C. Blood agar and MacConkey agar (Hi-Media, Mumbai India) were used as culture media to isolate the bacterial colonies. The isolated colonies were then identified by Gram stain and standard biochemical tests. Antibiotic susceptibility testing was performed by Kirby Bauer's disk diffusion method and interpreted as per Clinical and Laboratory Standards Institute guidelines.[4] Routine antimicrobial sensitivity tests were put for the following antibiotics.

Drugs on Gram-Negative Bacilli (GNB)

Amoxicillin/Clavulanic acid (20/10 μg), Trimethoprim/Sulphamethoxazole [Cotrimoxazole] (1.25/23.75 μg), Ceftriaxone (30 μg), Cefotaxime (30 μg), Gentamicin (10 μg), Amikacin (30 μg), Piperacillin/Tazobactum (100/10 μg), Ciprofloxacin (5 μg), Levofloxacin (5 μg), Imipenem (10 μg), Meropenem (10 μg), Netilmicin (30 μg), Tobramycin (10 μg), and Colistin were used.

Drugs on Gram-Positive (GP) Bacteria

Cefoxitin (30 μg), Penicillin G (10 units), Clindamycin (2 ug), Erythromycin (15 ug), Trimethoprim/Sulfamethoxazole (1.25/23.75 ug), Vancomycin (30 μg), Linezolid (30 μg), Teicoplanin (30 μg), Gentamicin (10 μg), and high-level Gentamicin (120 μg) were used.

Control strains were used for Kirby Bauer method:

- *Escherichia coli (*ATCC 25922)
- *Staphylococcus aureus (*ATCC 25923)
- *Pseudomonas aeruginosa (*ATCC 27853).

Statistical Analysis

Data were entered in excel sheet to prepare a master chart. Results were analyzed by counts and percentages using statistical methods.

Results

A total of 3,703 fluid samples were received from January 2018 to December 2018. Majority of them were ascitic fluid (49.9%) followed by pleural fluid (39.4%). Others (10.7%) were pericardial fluid, synovial fluid, bile, peritoneal dialysis fluid, bronchoalveolar lavage fluid, etc. Pathogenic bacterial growth was shown in 479 samples with an isolation rate of 12.93%.

Out of these 479 culture positive samples Gram-negative bacteria (GNB; 77.45%) were most commonly isolated than Gram-positive cocci (GPC; 22.55%). Predominant organisms were *E. coli* (31%) followed by *Acinetobacter* species (spp.) (20%), *Klebsiella* spp. (16%), *S. aureus* (11%), *Enterococcus* spp. (10%), *and Pseudomonas* spp. including *Aeruginosa* (9.8%). Less commonly isolated were *Streptococcus pneumoniae* (1.4%), *Proteus* spp. (0.6%), and *Citrobacter* spp. (0.2%) [Figure 1].

E. coli (40%) was the most commonly isolated organism in ascitic fluids followed by *Klebsiella* spp. (15%) and *Acinetobacter* spp. (13.8%). *Enterococcus* spp. (12%) was the most common Gram-positive isolate in ascetic fluid. In pleural fluids, *Acinetobacter* spp. (24.2%) was the most commonly isolated organism followed by *S. aureus* (21.8%). *E. coli* (14.5%) and *Klebsiella* spp. (14.5%) were other important isolates in pleural fluids. Bacteriological profiles of different body fluids are given in Table 1.

Out of total 3,703 fluid samples received; most of them were from IPD (66.3%) followed by emergency ward (30.7%) and then ICU (2.9%). However, ICU accounts for the most culture positivity rate (41.7%) followed by IPD (14%) and emergency ward (7.6%). Growth pattern of different body fluids from different locations is given Table 2 and Figure 2.

Gram-negative isolates are 100% sensitive to colistin, followed by imipenem and gentamicin.

Increased resistance shown toward third generation cephalosporins, amoxicillin + clavulinic acid, and piperacillin + tazobactum [Figures 3-6].

However, cotrimoxazole showed varied susceptibility – more sensitive in case of *Acinetobacter* than *E. coli. Klebsiella spp*. was the most resistant bacteria in our hospital among all the gram-negative isolates, while *Pseudomonas* showed mostly sensitive pattern with the most of the antibiotics tested.

Enterococcus spp. was the most resistant among Gram-positive isolates. They are only 35.4% sensitive to high level gentamicin but 100% sensitive to linezolid. The rate of isolation of vancomycin resistant enterococci in sterile body fluid was 9.5% in our hospital. *S. aureus* was 100% sensitive to vancomycin, linezolid,

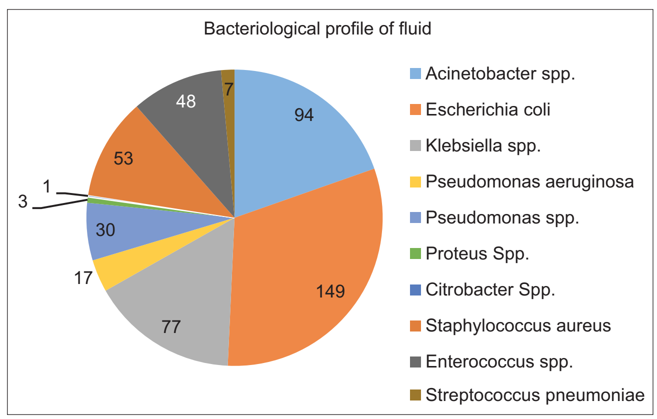

Figure 1: Total bacteriological profile

Table 1: Bacteriological profile of different body fluids

Organism	*Total no.*	*Ascitic Fluid*	*Pleural Fluid*	*Others*
Escherichia coli	149	90	18	41
Acinetobacter spp.	94	31	30	33
Klebsiella spp.	77	34	18	25
Pseudomonas spp.	30	17	6	7
Pseudomonas aeruginosa	17	5	8	4
Proteus spp.	3	-	-	3
Citrobacter spp.	1	-	-	1
Staphylococcus aureus	53	17	27	9
Enterococcus spp.	48	27	14	7
Streptococcus pneumonia	7	4	3	-

Table 2: Growth pattern of different body fluids from different locations

Sample	*EM*		*IPD*		*ICU*		*Total*	
	n (Positive)	*%*	*n (Positive)*	*%*	*n (Positive)*	*%*	*n (Positive)*	*%*
Ascitic Fluid	668 (63)	9.4	1157 (143)	12.3	24 (19)	79.1	1849 (225)	12.2
Pleural Fluid	447 (18)	4.0	959 (91)	9.4	52 (15)	28.8	1458 (124)	8.5
Others	23 (6)	26.1	341 (113)	33.1	32 (11)	34.3	396 (130)	32.8
Total	1138 (87)	7.6	2457 (347)	14.1	108 (45)	41.6	3703 (479)	12.9

EM: Emergency ward, IPD: Indoor patient department, ICU: Intensive care unit, *n*: Total no of samples

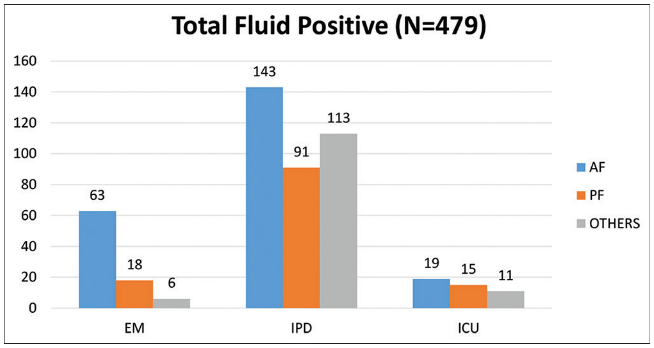

Figure 2: Distribution of growth pattern of various body fluids at different location. EM: Emergency ward, IPD: Indoor patient department, and ICU: Intensive care unit

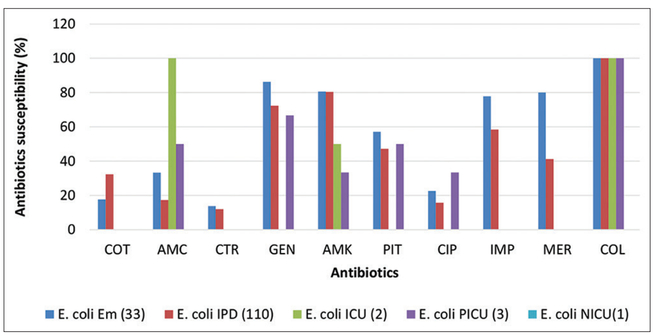

Figure 3: Antimicrobial sensitivity pattern (%) of Escherichia coli from different locations. COT: Cotrimoxazole, AMC: Amoxicillin+Clavulinic Acid, CTR: Ceftriaxone, GEN: Gentamicin, AMK: Amikacin, PIT: Piperacillin+Tazobactum, CIP: Ciprofloxacin, IMP: Imepenem, MER: Meropenem, and COL: Colistin

and teicoplanin, while they showed good sensitivity toward clindamycin but increased resistance toward cotrimoxazole and erythromycin [Figure 7].

Methicillin resistant *S. aureus* (MRSA) were 15.6% in our setting which was having similar susceptibility pattern as of MSSA. *S. aureus* isolated from IPD was found to be 33.4 % susceptible to penicillin G.

Discussion

Infection of sterile body fluid can lead to severe morbidity and mortality. The microorganisms as well as their antibiotic resistance patterns may change from time to time and place to place. The rapid development of multidrug resistance has complicated the treatment, for which we require a periodic monitoring of body fluid so that appropriate choice of drug can limit such infections.[5]

In our study, pathogen isolation rate was 12.93% which is in accordance to the studies by Rouf and Nazir[6] and Kasana *et al*.[7]

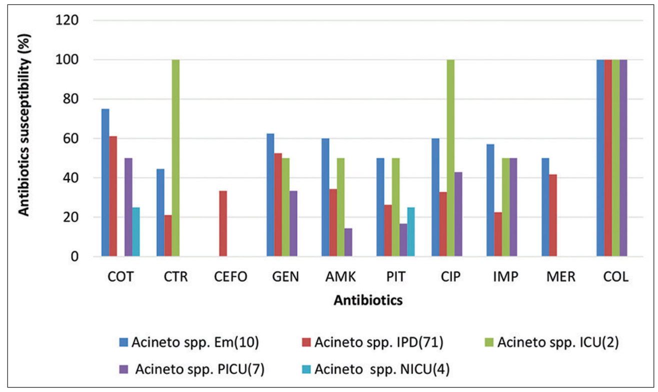

Figure 4: Antimicrobial sensitivity pattern (%) of *Acinetobacter* spp. from different locations. COT: Cotrimoxazole, CTR: Ceftriaxone, CEFO: Cefotaxime, GEN: Gentamicin, AMK: Amikacin; PIT: Piperacillin+ Tazobactum; CIP: Ciprofloxacin; IMP: Imepenem; MER: Meropenem; and COL: Colistin

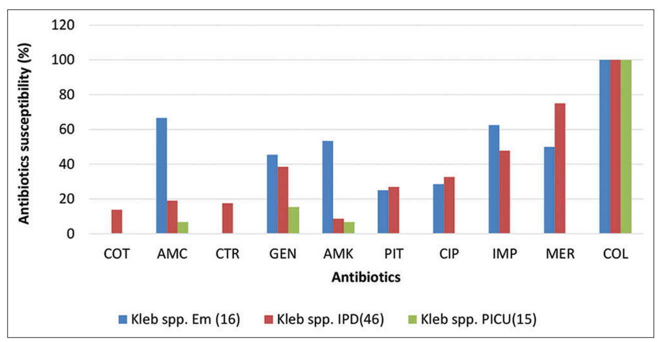

Figure 5: Antimicrobial sensitivity pattern (%) of *Klebsiella* spp. from different locations. COT: Cotrimoxazole, AMC: Amoxicillin+Clavulinic Acid, CTR: Ceftriaxone, GEN: Gentamicin, AMK: Amikacin, PIT: Piperacillin+Tazobactum, CIP: Ciprofloxacin, IMP: Imepenem, MER: Meropenem, and COL: Colistin

that showed an isolation rate of 10.8 % and 14.8%, respectively. However, the isolation rates were lesser in comparison to other studies on body fluids which reported 18.36%, 20.55%, and 22%, positivity.[8-10] Several studies done on body fluid profile showed discordant results in the spectrum of pathogens causing infection which may be due to indiscriminate use of antibiotics, patient specific factors such as surgical procedures, trauma or any other underlying conditions or by methodological factors such as proper specimen collection, transport, and culture.[10]

GNBs were predominated (77.45%) over gram-positives (22.55%) in our study. This predominance is in concordance with the findings of the similar study conducted by Rouf and

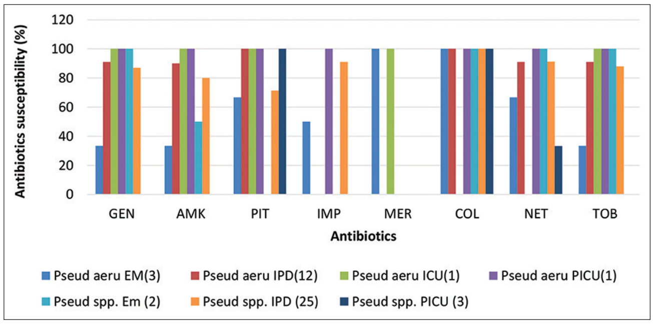

Figure 6: Antimicrobial sensitivity pattern (%) of *Pseudomonas* spp. from different locations. GEN: Gentamicin, AMK: Amikacin, PIT: Piperacillin+Tazobactum, IMP: Imepenem, MER: Meropenem, COL: Colistin, NET: Netilmicin, and TOB: Tobramycin

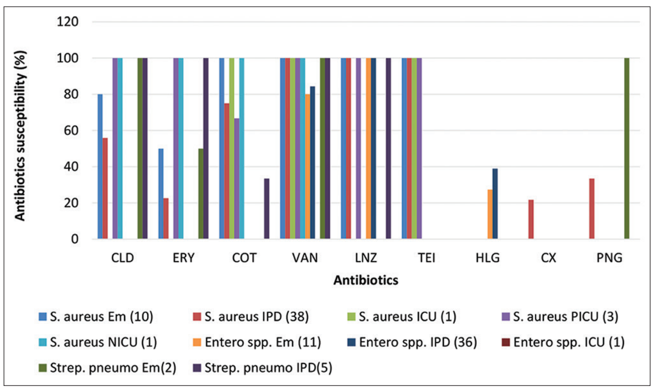

Figure 7: Antimicrobial sensitivity pattern (%) of Gram Positive bacterial isolates from different locations. CLD: Clindamycin, ERY: Erythromycin, COT: Cotrimoxazole, VAN: Vancomycin, LNZ: Linezolid, TEI: Teicoplanin, HLG: High Level Gentamicin, CX: Cefoxitin, and PNG: Penicillin G

Nazir.[6] that showed 70% GNBs and 30% gram-positives. Harshika *et al*.[3] and Vijaya and Anuradha[9] also showed similar GNB predominance. This may be due to their wide prevalence in the hospital environment[11,12] and frequent resistance to antibiotics may be the reason of their persistence and spread.[12]

In our study, *E. coli* (31%) was the most common isolate followed by *Acinetobacter* spp. (20%) among GNB, while *S. aureus* (11%) was most commonly isolated among GP. Our study findings are supported by various other studies such as Deb *et al*.[13] and Harshika *et al*.[3] However, this is in contrast to the study conducted by Sharma *et al*.,[10] where *Acinetobacter* spp. was the main isolate.

In ascitic fluid, *E. coli* was the most common bacteria found followed by *Klebsiella* spp. and *Acinetobacter* spp. which is similar to studies done by Sujatha *et al*.[14] and Vijaya and Anuradha.[9] *Enterococcus* spp. was the most common GP isolate in ascitic fluid in our study. This was in contrast to other studies[9,14] that shown *S. aureus which* is the most common isolate among GP in ascitic fluid. Our study is in accordance to study by Shivani *et al*.[15] that reported *Enterococcus* as the most common GP isolates in peritoneal fluid. Ding *et al*.[16] also shown that *Enterococcus spp.* is the predominant gram-positive organisms in spontaneous bacterial peritonitis. Researchers have shown that GNBs are predominant in the community-acquired infections, whereas Gram-positive organisms are predominant in the nosocomial infections.[16,17]

Among pleural fluid isolates, most common bacteria were *Acinetobacter* spp. followed by *S. aureus. E. coli* and *Klebsiella* spp. were other isolates. Our findings are in association with Sharma *et al*.,[10] who found *Acinetobacter* spp. and *E. coli* as the most common Gram-negative isolates in pleural fluid, while other study[9] showed that *Klebsiella* spp. and *E. coli* are the most common isolates in pleural fluids. Study by Vishalakshi *et al*.[18] showed *S. aureus* as the most common isolates from pleural fluid. In our study, *S. aureus* is the second most common isolates after *Acinetobacter spp.* and most common GP isolate in pleural fluid. This was supported by various studies[3,9,14] that showed *S. aureus* to be the most common GP isolate in pleural fluid which is the leading cause of empyema along with Gram-negative isolates. The isolation of aerobic Gram-negative or multiple pathogens from pleural fluid is associated with a poor prognosis and indicates a more aggressive antimicrobial chemotherapy in contrast to the empyema caused by Gram-positive pathogens.[6]

In this study, majority of the fluid received were ascetic fluid, also having higher isolation rates among all other fluids which are similar to study by Sharma *et al*.[10] Some other studies showed pleural fluid as the most common fluid received.[3,9] Although IPD accounted for most of the fluid received from, ICU had highest culture positivity rates. This may be due to more patient load in IPD, while more sick and infectious patient is in ICU.

In this study, most effective antibiotic against Gram-negative bacteria is colistin which is 100% sensitive. This is similar to the study of Sharma *et al*.[10] who reported 100% colistin sensitivity. Carbapenem showed fairly good sensitivity to GNB. This is in contrast to other studies by Harshika *et al*. and Tullu *et al*. that showed 100% carbapenemase sensitivity. Good sensitivity against gentamicin and increased resistant against cephalosporin and amoxicillin + clavulinic acid is also shown in studies of Harshika *et al*., Tullu *et al*., and Sharma *et al*. *Klebsiella* shown most drug resistant pattern in our study similar to the study by Vijaya and Anuradha *et al*. Good sensitivity pattern to available antibiotics among *Pseudomonas* is similar to several other studies.

Enterococci are highly resistant among Gram-positive organisms in our study. This is similar to the study of Vijaya and Anuradha[9] and Sharma *et al*.,[10] where *Enterococci* were showed increased resistance to gentamicin, tetracycline, and ciprofloxacin and highly sensitive to vancomycin and linezolid. *Enterococci* are regarded as hospital acquired pathogens that have become increasingly resistant to the available antibiotics and causing high mortality. Our study is in contrast to Harshika *et al*.[3] who showed 100% sensitivity of *Enterococcus* spp. to all antibiotics.

In our study, *S. aureus* is 100% sensitive to vancomycin, linezolid and teicoplanin such as other studies.[3,9,19] MRSA isolation rate is 15% in our study which is similar to the study of Vijaya and Anuradha[9] (17.66%). Mandira *et al*.[8] and Sharma *et al*.[10] have reported higher MRSA (28.57% and 38.5%), respectively. Variation in prevalence might be due to difference in efficacy of infection control practices, antibiotic usages, and health-care facility that differ in different hospital settings.

Conclusion

In recent years, rapid rise in bacterial resistance particularly in sterile body fluid infection caused significant morbidity and mortality. Regular surveillance of hospital associated infection and monitoring of antibiotic susceptibility pattern is required to reduce antimicrobial resistance. Culture antibiograms are the most important step of this process which is needed to form antibiotic policy of the hospital and choosing appropriate antibiotic therapy. A constant evaluation of trends in resistance of antimicrobials and antibiotic consumption patterns is essential for the judicious use of antibiotics and thus reducing development of multidrug resistance.

Acknowledgment

Authors acknowledge the immense help received from the scholars/authors/editors/publishers, whose articles are cited and included in references of this manuscript.

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Conflicts of Interest

The authors declare no potential conflicts of interest with respect to research, authorship, and/or publication of this article.

References

1. Daur AV, Klimak F, Cogo LL, Botao GD, Monteiro CL. Enrichment methodology to increase the positivity of cultures from body fluids. Braz J Infect Dis 2006;10:372-3.
2. Fridkin SK. Increasing prevalence of antimicrobial resistance in intensive care units. Crit Care Med 2001;29:N64-8.
3. Harshika YK, Shobha MK, Patil AB, Smita NR. A study on bacteriological profile and antimicrobial resistance pattern from various body fluids of patients attending the tertiary care Hospital, KIMS, Hubli. Indian J Microbiol Res 2018;5:530-4.
4. Clinical Laboratory Standard Institute. 2017. Performance Standard for Antimicrobial Disk Susceptibility Tests. Approved Standard. 20th ed. Supplement/M2- A92006:26(1). Clinical Laboratory Standard Institute; 2017.
5. Chauhan M, Manish S, Mahajan S. Aerobic Bacterial Profile and antibiotic sensitivity pattern of pus isolates in a tertiary care Hospital. Int J Curr Microbiol Appl Sci 2015;4:784-7.
6. Rouf M, Nazir A. Aerobic bacteriological profile and antimicrobial sensitivity pattern of bacteria isolated from sterile body fluids: A study from a tertiary care hospital in North India. Micrbiol Res J Int 2019;28:1-10.
7. Kasana D, Purohit G, Nair D. Bacteriological profile and antibiogram in various body fluids in a tertiary care hospital in north India: A 6 years observational study. Int J Al Recent Trends Sci Technol 2015;16:432-5.
8. Mandira R, Annie B, Elantamilan D. A study on bacteriological profile and antibiogram in sterile body fluids from a tertiary care hospital in Northeast India. Int J Sci Res 2018;7:42-8.
9. Vijaya DS, Anuradha B. An aerobic bacteriological profile and antibiogram of various body fluids from a tertiary care hospital in Telangana, India a 5 year study. Int J Curr Microbiol Appl Sci 2019;8:592-601.
10. Sharma R, Anuradha, Nandini D. Bacteriological profile and antimicrobial sensitivity pattern in sterile body fluids from a tertiary care hospital. J Appl Microbiol Biochem 2017;1:1.
11. Qadeer A, Akhtar A, Ul Ain Q, Saadat S, Mansoor S, Assad S, *et al*. Antibiogram of medical intensive care unit at tertiary care hospital setting of Pakistan. Cureus 2016;8:e809.
12. Al-Jawady Z, Al-Habib HM. Antibiogram profiles of bacterial isolates from intensive care units in Mosul teaching hospitals. Rafidain J Sci 2012;23:52-9.
13. Deb A, Mudshinkar S, Dohe V, Bharadwaj R. Bacteriology of body fluids with an evaluation of enrichment technique to increase culture positivity. J Evol Med Dent Sci 2014;3:15-23.
14. Sujatha R, Pal N, Arunagiri D, Narendram D. Bacteriological profile and antibiotic sensitivity pattern from various body fluids of patients attending Rama Medical College hospital Kanpur. Int J Adv Case Rep 2015;2:119-24.
15. Khullar S, Rathore L, Khatri PK, Parihar RS, Meena S, Bora A, *et al*. Identification and antibiogram of various gram positive bacterial isolates from pyogenic samples by VITEK® 2 compact system. Int J Curr Microbiol Appl Sci 2016;5:66-77.
16. Ding X, Yu Y, Chen M, Wan C, Kang Y, Lou J. Causative agents and outcome of spontaneous bacterial peritonitis in cirrhotic patients: Community-acquired versus nosocomial infections. BMC Infect Dis 2019;19:46-53.
17. Shi L, Wu D, Wei L, Liu S, Zhao P, Tu B, *et al*. Nosocomial and community-acquired spontaneous bacterial peritonitis in patients with liver cirrhosis in China: Comparative microbiology and therapeutic implications. Sci Rep 2017;7:4602-5.
18. Vishalakshi B, Hanumanthappa P, Krishna S. A study on aerobic bacteriological profile of sterile body fluids. Int J Curr Microbiol Appl Sci 2016;5:120-6.
19. Kumar S, Joseph NM, Easow JM, Singh R, Umadevi S, Pramodhini S, *et al*. Prevalence and current antibiogram of staphylococci isolated from various clinical specimens in a tertiary care hospital in Pondicherry. Intern J Microbiol 2012;10. Available from: https://ispub.com/IJMB/10/1/13757