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Original Research Article

Study of the antibiotic sensitivity pattern of bloodstream infections in gynaecological ICU

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ABSTRACT

Background: Severe sepsis is one of the leading causes of death even in the developed nation, among critically ill patients admitted in intensive care units (ICU). Decreasing antibiotic susceptibility with increasing length of hospital stay increases the burden on the healthcare system.
Methods: A total of 75 samples taken from the patients suspected with bloodstream infection in the gynaecological ICU were processed as per standard protocol. Identification of bacteria was carried out with the help of relevant biochemical tests. Antibiotic susceptibility testing was done by Kirby-Bauer Disk diffusion method according to CLSI guidelines.
Results: Positive blood culture was seen in 21 (28%) samples. Out of these 13 (61.90%) were gram negative bacteria and 8 (38.09%) was gram positive cocci. *Escherichia* spp., *Klebsiella* spp. and *Acinetobacter* spp. were the predominant isolates in gram negative bacteria. Gentamicin, Piperacillin-Tazobactam, Imipenem and Levofloxacin were the most sensitive antibiotics while Cefazolin, Cefuroxime, Cefepime and Ceftazidime were the most resistant antibiotics.
Conclusions: Gram negative bacteria are an emerging cause of sepsis in ICU patients while gram positive bacteria still remain prevalent. The purpose of this study is to identify the bacterial cause of septicaemia in general intensive care unit (GICU) patients.

Keywords: Microbiological profile, Sepsis, ICU, Bloodstream infections, Antibiotic sensitivity pattern

INTRODUCTION

Severe sepsis is one of the leading cause of death even in the developed nation among critically ill patients admitted in intensive care units (ICU) other than cardiac cause.[1] Bacteraemia is a state in which bacteria circulate through vascular system whereas Septicaemia is a life threatening condition when bacteria multiply at a rate that exceeds their removal by phagocytes.[4] The symptoms are produced by microbial toxins and cytokines produced by inflammatory cells.[4] Patients admitted to the critical care units of the hospitals are always at a higher risk of developing nosocomial BSIs which results in high morbidity and mortality among these patients. Currently, sepsis/septic shock and associated bloodstream infections (BSI) are among the most prevalent causes of morbidity and mortality in many European and North American countries with an estimated 157,000 deaths annually in Europe and as much as 94,000 in North America.[3] Organism isolated from blood culture vary according to geographical distribution, and development of multidrug resistant organism is of great concern, as they prolong hospital stay, increase cost of treatment and can be a cause of high mortality.[4] In a study conducted in an European teaching hospital in 2018 a total, 3,349 (86.1%) blood cultures were negative and 541 (13.9%) were positive for one or more microorganisms.[3] Decreasing antibiotic susceptibility with increasing length of hospital

stay has previously been illustrated for the infection of selected organ systems.[6] From 2008 to 2017, a study conducted in the department of infectious diseases in Switzerland concluded that after excluding possible skin contaminants, they observed 3788 bacteraemias and 130 fungaemias out of 6506 ICU-BSIs.[6] Main research topics include faster detection of causative microorganisms, development of novel treatment strategies that save lung and kidney function, and development of more individualized treatment approaches.[11] Currently, antibiotics and antibiotic-resistance genes have been reported in surface water, effluents from sewage treatment plants, soils, and animal wastes.[8] Owing to this wide distribution, WHO declared it as a serious public health crisis of the 21st century.[8]

The purpose of this study is to identify the bacterial cause of septicaemia in GICU patients in order to provide a timely and fairly accurate diagnosis for better treatment of patients while also analysing and ensuring the judicious use of antimicrobial therapy.

METHODS

A prospective study was conducted in Department of Microbiology, Netaji Subhash Chandra Bose Medical College, Jabalpur from 7th April 2021 to 20th January 2022. Ethical approval for this study wasn't required. All patients in the gynaecological ICU suspected of sepsis were included in the study. Exclusion criteria included patients less than 18 years of age. A total of 75 blood samples were received from the gynaecological ICU. Samples were collected under aseptic precautions with standard method and transferred to previously prepared blood culture bottles. Bottles were correctly labeled and transported to the bacteriology section of the Department of Microbiology with minimal delay. The blood culture bottles were incubated overnight at 37°C and then sub-cultured on to nutrient agar, blood agar, chocolate agar and MacConkey agar to look for any growth. Any growth that was observed after overnight incubation at 37°C was identified with the help of colony morphology, gram staining and relevant standard biochemical test such as catalase, coagulase, tsi, oxidase, citrate, urease, methyl red, indole, vogues Proskauer. Antimicrobial susceptibility tests were done in Muller Hinton Agar by the Kirby Bauer Disc diffusion method as per CLSI guidelines.[2]

Blood culture bottles which showed no signs of any growth after 5 days of incubation (growth on MacConkey/ blood agar/ haemolysis/ turbidity) were reported as negative after a final confirmatory subculture.

RESULTS

During the period from April 2021 to January 2022, a total of 75 samples were received from septicaemia suspected patients from the gynaecological ICU and processed routinely. Out of 75 blood cultures, 21 (28%) were found to be positive for culture growth (Table 1).

Table 1: Distribution according to positive cultures.

Culture status	Samples (%)
Positive	21 (28)
Negative	54 (72)
Total	75 (100)

In the age distribution of the patients it was observed that majority of the patients belonged to the age group of 18-22 (26.66%). The number decreased from 58-72 years with average of 1 case (1.33%) (Table 2).

Table 2: Age distribution of cases.

Age distribution (years)	No. of cases (%)
18-22	20 (26.66)
23-27	16 (21.33)
28-32	14 (18.66)
33-37	10 (13.33)
38-42	5 (6.66)
43-47	4 (5.33)
48-52	1 (1.33)
53-57	3 (4)
58-62	1 (1.33)
63-67	0 (0)
68-72	1 (1.33)
Total	75 (100)

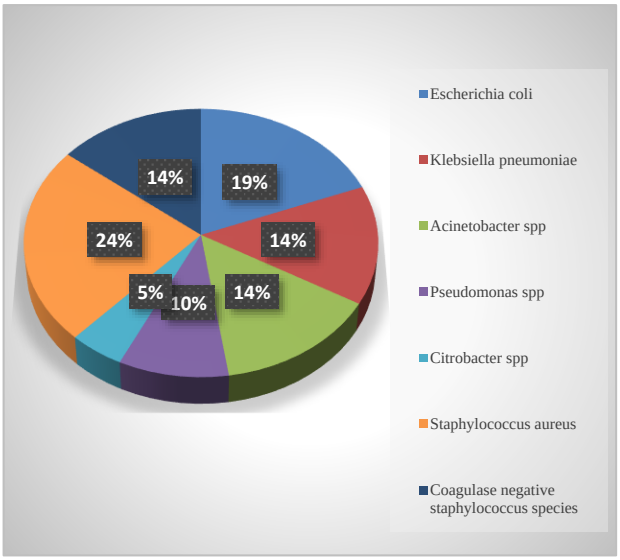

Figure 1: Distribution of bacteria isolated in blood culture samples of patients admitted in the gynaecological ICU. *Staphylococcus aureus* was the most common isolate followed by *Escherichia coli* (19%), *Klebsiella pnuemoniae* (14%), *Acinetobacter* spp. (14%), Coagulase negative *Staphylococcus* spp. (14%), *Pseudmonas* spp. (10%) and *Citrobacter* spp. (5%).

Among the 21 positive cultures, gram negative and gram positive constituted 13 (61.90%) and 8 (38.09%) respectively. Out of the gram negative bacteria, *Escherichia coli* 4 (25%) was the predominant isolate followed by *Klebsiella pneumoniae* 3 (14.28%), *Acinetobacter* spp. 3 (14.28%), *Pseudomonas* spp. 2 (12.5%), *Citrobacter* spp. 1 (4.76%) and *Staphylococcus aureus* 5 (23.80%) and coagulase negative *Staphylococcus* spp. (CONS) 3 (14.28%) in gram positive isolates (Figure 1).

Among the gram negative isolates, *Escherichia coli* showed least resistance to Imipenem and Doxycycline. *Klebsiella pneumoniae* showed least resistance to Cefotaxime-Clavulanic acid, Piperacillin-tazobactam, Imipenem, Doxycycline and Levofloxacin. Citrobacter spp showed least resistance to Gentamicin, Cefotaxime-Clavulanic acid, Amoxycillin-Clavulanic acid, Piperacillin-tazobactam, Cefotaxime, Doxycycline, Imipenem, Levofloxacin whereas Acinetobacter spp. showed least resistance to Piperacillin-tazobactam, Doxycycline and Imipenem (Table 3).

Among the gram positive isolates, staphylococcus aureus and coagulase negative *Staphylococcus* spp. (CONS) both showed least resistance to Doxycycline, Gentamicin, Levofloxacin and Vancomycin (Table 4).

Table 3: Antibiotic sensitivity pattern of gram negative isolates of sepsis patients.

S. no.	Antibiotics	Percentage of sensitive strains				
		E. coli (n=4)	***Klebsiella pneumoniae* (n=3)**	***Acinetobacter* (n=3)**	***Pseudomonas* spp. (n=2)**	***Citrobacter* (n=1)**
1	Gentamicin	50	33.33	33.33	50	100
2	Cefazolin	25	0	33.33	0	0
3	Cefotxime+Clavulanic acid	25	66.66	66.66	50	100
4	Amoxycillin-Clavulanic acid	50	33.33	0	NT	100
5	Piperacillin-Tazobactam	50	33.33	100	50	100
6	Cefuroxime	0	0	33.33	0	0
7	Cefotaxime	25	0	0	50	100
8	Doxycycline	75	66.66	100	100	100
9	Imipenem	100	66.66	100	0	100
10	Cefepime	0	33.33	0	0	0
11	Ceftazidime	0	0	0	0	0
12	Levofloxacin	50	66.66	33.33	50	100
13	Cotrimoxazole	50	33.33	33.33	0	100
14	Tobramycin	NT	NT	NT	100	NT

Table 4. Antibiotic sensitivity pattern of gram positive isolates of sepsis patients

S. no.	Antibiotics	Percentage of sensitive strains	
		***Staphylococcus aureus* (n=5)**	**Coagulase negative *Staphylococcus* spp. (n=3)**
1	Amoxycillin-Clavulanic acid	40	0
2	Azithromycin	40	33.33
3	Cefotaxime	40	66.66
4	Ciprofloxacin	60	66.66
5	Doxycycline	80	100
6	Erythromycin	20	33.33
7	Gentamicin	80	100
8	Levofloxacin	80	100
9	Vancomycin	100	100
10	Penicillin	20	0

DISCUSSION

In our study, culture positivity was found to be 28%. The rate of culture positivity in septicaemia cases nearly similar to our study were reported in the study of Wasihun et al (28%), Agrawal R, Ranjan K (26.69%) and Gill MK, Sharma S (24.8%).[7,9] This was not in concordance with the study of Gohel et al (9.2%) and Wu et al (4.57%).[5,8] Slight variation may be due to many factors like geographical locations, patient type, timing and number of blood cultures or difference in blood culture system.[9]

In the present study, 61.09% infections were caused by gram negative bacteria and 38.09% were due to gram

positive bacteria. This was found to be similar to the study of Agrawal R, Ranjan K, who concluded that in positive samples gram negative bacteria and gram positive bacteria were 68.35% and 31.65% respectively.

In our study out of these, *Escherichia* spp. was the most common amongst gram negative bacteria and *Staphylococcus aureus* amongst gram positive bacteria. Wasihun et al found the predominant isolate to be *Staphylococcus aureus* (10.3%), CONS (8.5%), *E. coli* (3.1%), *Citrobacter* spp. (1.7%) and *S. typhi* (1.6%) which was similar to study by Gill MK, Sharma S.[7,9] who reported as 53% gram positive bacteria followed by 39.3% of gram Negative bacteria and 7.9% of non-albicans *Candida*.

We observed in our study that Imipenem and Doxycycline (66.66-100%) were most sensitive for gram negative bacteria. This was comparable with as shown by Agrawal R, Ranjan K that Imipenem and Linezolid were most sensitive for gram negative bacteria. Similarly, in comparison, Wasihun et al also had *E. coli* showing 60% resistance to Ceftriaxone, while Acinetobacter showed 80% and 60% resistance to Ceftriaxone and Cotrimoxazole respectively.[7] In our study, gram positive bacteria were most sensitive to Vancomycin, Levofloxacin, Gentamicin, and Doxycycline. Cefazolin, Cefuroxime, Cefepime and Ceftazidime showed maximum resistance among gram negative isolates. Gram positive bacteria showed maximum resistance to Penicillin, Erythromycin and Cefuroxime.

In this study upon testing for antimicrobial susceptibility testing it was seen that, *Escherichia coli* showed high sensitivity for Doxycycline (75%) Imipenem (100%) followed by Amoxycillin Clavulanic acid (50%) and was most resistant to Cefuroxime (100%), Ceftazidime (100%) and Cefepime (100%). Study by Agrawal R, Ranjan K showed that *Escherichia coli* was most sensitive to Doxycycline (76.92%) and Imipenem (92.31%) while it was most resistant to Ceftriaxone (69.23%) and Ceftazidime (61.34%). Wasihun et al concluded in their study that Escherichia coli was sensitive to Doxycycline (60%).[7] So present study showed comparable sensitivity for Doxycycline while higher sensitivity for Imipenem in *E. coli* than that of Agrawal R, Ranjan K.[1]

In the present study, *Klebsiella pneumoniae* was found to be most sensitive to Cefotaxime Clavulanic Acid (66.66%), Doxycycline (66.66%) and Imipenem (66.66%). Study by Sonawane et al exhibited that Klebsiella spp showed a high degree of resistance to Ceftazidime Clavulanic acid combination (7.81%) and highest sensitivity for Imipenem (95.31%).[4] In the study by Agrawal R, Ranjan K, *Klebsiella pneumoniae* was highly sensitive for Imipenem (75%) followed by Doxycycline (66.67%). So results of present study in Doxycycline susceptibility were comparable with that of Agrawal R, Ranjan K.[1] The present study showed higher sensitivity for Cefotaxime Clavulanic acid, while a lower sensitivity was observed for Imipenem compared to Sonawane.[4]

In the present study, *Pseudomonas* spp. was most sensitive to Imipenem (100%) and Tobramycin (100%) while it was most resistant to Cefotaxime (50%) and Piperacillin tazobactam (50%). Sonawane concluded in their study that *Pseudomonas aeruginosa* showed highest sensitivity for Imipenem (95%) followed by Piperacillin tazobactam while it showed a lower sensitivity to Tobramycin (50%), Ceftazidime (48.33%).[4] So the present study was comparable to that of Sonawane for sensitivity of Imipenem and Cefotaxime.[4] Results of Tobramycin sensitivity were higher in comparison.

In the present study, Citrobacter spp showed highest sensitivity to most drugs like Gentamicin (100%), Cefotaxime Clavulanic acid (100%), Amoxycillin Clavulanic acid (100%), Piperacillin Tazobactam (100%), Cefotaxime (100%), Doxycycline (100%), Imipenem (100%), Levofloxacin and Cotrimoxazole (100%).

In contrast, study by Wasihun et al showed that Citrobacter spp. was resistant to Amoxycillin Clavulanic acid (50%), Gentamicin (25%), Doxycycline (50%), Cotrimoxazole (37.5%).[7] Study by Agrawal R, Ranjan K showed that *Citrobacter* spp was most sensitive to Cefoperazone sulbactam (100%), Doxycycline (100%), Levofloxacin (100%), Piperacillin tazobactam (100%) and Imipenem (100%). Thus, the present study was comparable with that of Agrawal R, Ranjan K while, the study showed higher sensitivity compared to that of Wasihun et al.[1,7]

In the present study, *Acinetobacter* spp. was the most sensitive to Piperacillin Tazobactam (100%), Doxycycline (100%), Imipenem (100%). In the study by Sonawane, *Acinetobacter* spp. was most sensitive for Imipenem (88%) followed by Piperacillin Tazobactam (54%).[4] Sensitivity to Imipenem was comparable to the study by Sonawane and Agrawal R, Ranjan K who demonstrated a sensitivity of 100%.[1,4] In the present study, Piperacillin tazobactam showed higher sensitivity values compared to Sonawane.[4]

In the present study, *Staphylococcus aureus* was observed to be most sensitive to Vancomycin (100%), Doxycycline (80%), Gentamicin (80%) and Levofloxacin (80%) while it was most resistant to Penicillin. Study by Sonawane demonstrated that Staphylococcus aureus was most sensitive to Vancomycin (100%) and Gentamicin (83.33%).[4] In a study by Gohel K, they concluded that Staphylococcus aureus showed sensitivity to Doxycycline (73%) and Vancomycin (78%).

The present study results for Vancomycin susceptibility were comparable to that by Sonawane J whereas,

Gentamicin and Doxycycline showed higher sensitivity comparatively.[4]

In the present study, coagulase negative *Staphylococcus* spp was highly sensitive to Vancomycin (100%), Doxycycline (100%), Gentamicin (100%) and Levofloxacin (100%). In the study by Wasihun et al CONs showed highest sensitivity to Gentamicin (81.9%).[7] In the study by Agrawal R, Ranjan K, CONS showed sensitivity to Vancomycin (100%), Gentamicin (83.33%) and Doxycycline (83.33%).[1] So resistance pattern for these drugs was comparable with the present study.

ICU-associated gram-negative bloodstream infection in a setting of limited treatment options can adversely impact outcomes.[10] Despite the recent decline in case-fatality rates, sepsis will remain a major health burden worldwide due to its increasing incidence and the emergence of antibiotic resistance.[11]

CONCLUSION

The present study concluded that gram negative bacteria are an emerging cause of infections in gynaecological patients while Gram positive bacteria, especially *Staphylococcus aureus* remains an important cause of BSI in patients in critical care units. It implies that blood cultures must always be done in all cases of suspected bacteraemia and septicaemia and once the sensitivity pattern of the isolate is known de-escalation of the high-end antimicrobials should be considered to reduce the antimicrobial pressure.

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