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Bacteriological profile of blood culture isolation and their antibiotic susceptibility pattern in BIMR Hospital, Gwalior, India

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Abstract: Blood stream infections can result in sepsis, a condition that can be fatal. Blood culture is the most effective way to diagnosis of sepsis. Multi-drug resistant (MDR) organisms have a higher probability to raise mortality risk of patients. Blood sample was collected and transferred into a bottle of blood culture and incubated for 5 days. The most typical organism isolated was *Stenotrophonomas maltophilia* (39.65%) which was followed by *Staphylococcus aureus* (20%) in terms of frequency. The most frequent isolate, *Stenotrophomonas maltophilia*, was 100% sensitive to Chloramphenicol, 95.65% to Trimethoprim-sulfamethoxazole and 86.95% sensitive to Levofloxacin. Daptomycin, vancomycin and linezolid showed 100% sensitivity against all identified gram positive bacilli (GPC). According to the current study, the most prevalent isolates from blood cultures were *stenotrophomonas maltophilia* and *staphylococcus aureus,* which is predominant organisms causing septicemia. *Stenotrophomonas maltophilia*, had greater susceptibility to chloramphenicol, trimethoprim-sulfamethoxazole and levofloxacin. *Staphylococcus aureus*, showed higher sensitivity to daptomycin, vancomycin, linezolid.

Keywords: multi-drug resistant; MDR; septicaemia; blood culture; blood-stream infections; *stenotrophomonas maltophilia*; antibiotic susceptibility.

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Pranshu Sharma is a post-graduate student of microbiology. He has a huge bent of mind towards research as well as teaching. He conducted the work under the supervision of Dr. Kumar and Dr. Anju Goyal. He has several publications in journals. Currently, he is working with a reputed multinational industry sharing his knowledge to the industry.

Anju Goyal is a Microbiologist as well as a medical practitioner. She has a teaching as well as research experience of more than tenyears. She has guided research students at UG, PG as well as PhD level. She has several research publications in national as well as international journals. Currently, she is working as a Medical Practitioner with BIMR Hospital, Gwalior sharing her experience with the hospital as well as curing patients.

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1 Introduction

A blood culture has a strong prognostic value, is a key element in the proper identification of blood stream infections, and is crucial for the following therapy of septicemia (Negussie et al., 2015; Buttery, 2002). Septicemia caused due to huge quantities of micro-organisms are found in the blood, which is potentially fatal condition that is also known as blood poisoning (Gyawali et al., 2019) Bacteria can enter in the

circulation during surgery and due to catheters and other foreign objects get entering in the arteries or veins. Sepsis is defined as systemic inflammatory response syndrome (SIRS) resulting from a suspected or proven infectious etiology. The clinical spectrum of sepsis begins when a systemic (e.g., bacteremia, fungemia, viremia) or localised (e.g., meningitis, pneumonia, pyelonephritis infection progresses from sepsis to severe sepsis. Further deterioration leads to septic shock, multiple organ dysfunction syndrome (MODS) and possibly death (Bone et al., 2009).

The development of antimicrobial agent resistance in micro-organisms, makes the sharp rise in septicemia rates in developing nations a significant public health issue that poses the largest difficulty for doctors in the choice of effective antimicrobial medicines. Numerous investigations carried out in India and many other nations across the world have identified an increase in antibiotic resistance among instances of septicemia (Mohanty et al., 2017). This study aims to identify micro-organisms early and evaluate their patterns of antibiotic susceptibility, In order to inform doctors about pathogens that might endanger for patients. The pattern of antibiotic susceptibility to common pathogens is evolving daily, therefore it's critical to have the most up-to-date information possible to influence local, empirical antibiotic selection.

2 Materials and methods

A total of 351 specimens were gathered from various IPD and OPD appointments across all hospital departments. A 5–10 ml blood specimen was drawn using aseptic methods. After that, the blood specimen is transferred into a blood culture bottle with soybean-casein digest broth and sodium polyenetholsulfonate (anticoagulant). The identification of the microorganisms using standard operational procedures. After five days of 37°C incubation with an automated blood culture system (BD BACTEC FX 40), the system was able to detect the presence of bacteria in the culture bottle. When the system indicated a positive blood culture, the growth of microorganisms was sub-cultured on Blood agar and MacConkey agar at 37°C overnight, and the gram-stain and biochemical test were performed to determine the growth of bacteria. To distinguish between the various kinds of gram-positive cocci (GPC) species, coagulase and catalase tests were used. For the identification and distinction of enterobacteriaceae species, the gram-negative bacilli (GNB), IMViC test was applied that includes the tests Indole, Methyl red, Voges-Prskauer, and Citrate. And triple sugar iron (TSI) was also used as additional test.

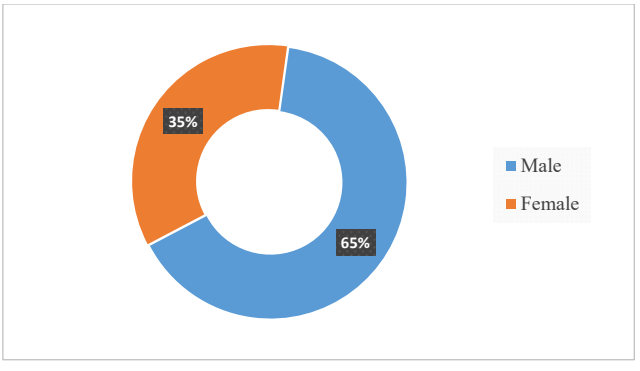
The Kirby-Bauer disc diffusion technique was used to assess the antibiotic susceptibility in accordance with the Clinical and Laboratory Standards Institute (CLSI) criteria. For this test, Mueller Hinton agar was utilised, and several antibiotics discs were put on the agar, including Amikacin (30 µg), gentamicin (30 µg), Ertapenem (10 µg), Imipenem (10 µg), Meropenem (10 µg), Cefazolin (30 µg), Cefuroxime (30 µg), Cefoxitin (30 µg), Ceftazidime (30 µg), Ceftriaxone (10 µg), Cefepime (30 µg), Ampicillin (10 µg), Ampicillin-Sulbactum (10 µg), Pipracillin-Tazobatam (10 µg), Ciprofloxacin (5 µg), Levofloxacin (5 µg), trimethoprim-sulfamethoxazole (25 µg), Ceftazidime-Avibactam (30 µg), Chloramphenicol (30 µg), Penicillin-G (2 units), Vancomycin (30 µg), Clindamycin (30 µg), Erythromycin (15 µg), Moxifloxacin (5 µg), Doxycycline (30µ g), Daptomycin, Trimethoprime-Sulfamethoxazole, Linezolid (30 µg), Rifampin (5 µg).

3 Results

351 blood samples in total were collected for this investigation, which was carried out between February 2022 and June 2022. Out of 351 blood samples tested throughout this investigation, 106 (30.1%) blood cultures proved to be positive. This study's high-risk age range was between 51 and 60 years (27.35%), and male accounting for 65% and female 35% (Figure 1).

GNB, GPC, and *candida* are present in relative amounts of 54.7%, 37.7%, and 7.5%.The most frequent isolate in GNB is Stenotrophomonas maltophilia, which accounts for 39.65% of the total. Other common isolates include *Acinetobacter* (13.79%), *Klebsiella* (12.06%), *Escherichia coli* (10.34%), *Salmonella-typhi* (6.89%), *Achromobacter spp.* (3.44%), *Proteus mirabilis* (3.44%), *Serretia marcescens* (3.44%), *Burkholderiacepacia* (1.72%), *Enterobacter cloacae* (1.72%), *Pantoea agglomerans* (1.72%) and *Pseudomonas aeruginosa* (1.72%) (Fig 2).37.7% of the isolates were gram-positive bacteria, including normal skin flora (60%) *Staphylococcus aureus* (20%) *Enterococcus spp.* (10%) *CONS* (7.5%), and *Streptococcus spp.* (2.5%) (Figure 3) Candida species of fungi were found (7.5%).

Figure 1 Genders distribution of positive cases (n = 106) (see online version for colours)



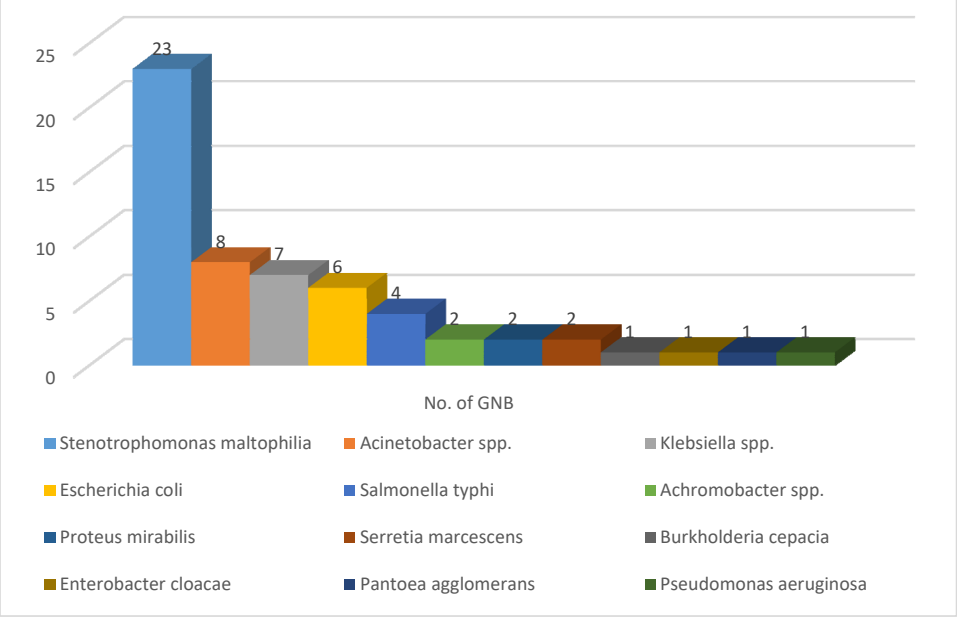
Gram-positive bacteria were completely susceptible to daptomycin, vancomycin, and linezolid, and staphylococcus aureus was also completely susceptible to these antibiotics. The most prevalent strain of gram-negative bacteria, *Stenotrophomonas maltophilia*, was completely susceptible to Chloramphenicol, 95.65% sensitive to Trimethoprim-sulfamethoxazole, and 86.95% sensitive to Levofloxacin. *Enterococcus spp.* are intrinsic resistance (IR) to cephalosporin, aminoglycosides, clindamycin and trimethoprime-sulfamethoxazole.

4 Discussion

An effort was undertaken in this study to analyses both bacterial profile and pattern of antibiotic sensitivity found in positive blood cultures. In hospitalised patients, blood stream infections are a significant source of morbidity and death. This study demonstrated that both gram-positive and gram-negative bacteria, the majority of which were multi-drug-resistant, were to blame for BSI.

The proportion of blood cultures that were positive in our research was 30.1% (106/351). The percentage of positive blood cultures in the study by Soni et al. (2019) was identical to our study results 28.09% (127/452). There was a male accounting for 65% and female 35% in this study (Figure 1). Gohel et al. (2014) revealed comparable findings, with males accounting for 67.43% and female accounted for 32.57%. In another study done by Mohanty et al. (2017) showed Male 66% and female 34%. Given that most patients in India receive antibiotics before being admitted to a tertiary care facility, variations in blood culture positive rates may result from this, as well as from self-medication given that medicines are widely accessible over the counter. Sincere attempts were undertaken to collect the blood samples prior to the administration of the antibiotics, which might have resulted in a high percentage of cultures that were positive.

Figure 2 Proportion of gram-negative organisms (see online version for colours)



In this study GNB 54.7% and GPC 37.7%. This is consistent with the research conducted by Ahmed and Hussain (2014), in which GNB accounted for 55.55% and GPC for 38.09%. Gram negative bacteremia was the most prevalent etiological agent (54.7%) with Stenotrophomonas maltophilia (39.65%) is the most common isolate. Isolated Gram-negative bacteria from BSI culture-positive cases including Acinetobacter (13.79%), Klebsiella (12.06%), Escherichia coli (10.34%), Salmonella-typhi (6.89%), Achromobacter spp. (3.44%), Proteus mirabilis (3.44%), Serretia marcescens (3.44%), Burkholderiacepacia (1.72%), Enterobacter cloacae (1.72%), Pantoea agglomerans (1.72%) and Pseudomonas aeruginosa (1.72%) (Figure 2). Because stenotrophomonas maltophilia is a free-living bacterium found in most aquatic and humid habitats, we saw its preponderance in our study (39.55%). It is not regarded as a typical component of human flora. It has been identified from a range of aquatic nosocomial sources and is often thought to be a low-virulence, opportunistic pathogen that may thrive on wet surfaces (such as exposure to nebulisers, endoscope, and shower heads) (Brooke, 2012).

Table 1 Antibiotic susceptibility pattern of GNB

ntibiotics	*Stenotrophomonas Maltophilia (N-23) (%)*	*Acinetobacter spp. (N-8) (%)*	*Klebsiella spp. (N-7) (%)*	*Escherichia coli (N-6) (%)*	*Salmonella typhi (N-4) (%)*	*Achromobacter spp. (N-2) (%)*	*Proteus mirabilis (N-2) (%)*	*Serretia marcescens (N-2) (%)*	*Burkholderia cepacia (N-1) (%)*	*Enterobacter cloacae (N-1) (%)*	*Pantoea agglomerans (N-1) (%)*	*Pseudomonas Aeruginosa (N-1) (%)*
mikacin	IR	1(12.5%)	1(14.28%)	6(100%)	(0%)	(0%)	1(50%)	(0%)	(0%)	(0%)	1(100%)	(0%)
entamycin	IR	1(12.5%)	1(14.28%)	4(66.6%)	(0%)	(0%)	1(50%)	(0%)	(0%)	1(100%)	1(100%)	(0%)
rtapenem	IR	IR	(0%)	4(66.6%)	4(100%)	(0%)	(0%)	(0%)	IR	(0%)	1(100%)	IR
nipenem	IR	(0%)	1(14.28%)	5(83.3%)	4(100%)	2(100%)	(0%)	(0%)	(0%)	(0%)	1(100%)	(0%)
[eropenem	IR	(0%)	1(14.28%)	5(88.3%)	4(100%)	(0%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	(0%)
efazolin	IR	IR	(0%)	1(16.6%)	(0%)	(0%)	(0%)	(0%)	IR	(0%)	1(100%)	IR
efuroxime	IR	IR	(0%)	1(16.6%)	(0%)	(0%)	(0%)	(0%)	IR	(0%)	1(100%)	IR
efoxitin	IR	IR	(0%)	2(33.3%)	(0%)	(0%)	(0%)	(0%)	IR	(0%)	1(100%)	IR
eftazidime	(0%)	(0%)	(0%)	2(33.3%)	3(75%)	2(100%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	(0%)
eftriaxone	IR	(0%)	(0%)	1(16.6%)	3(75%)	(0%)	(0%)	(0%)	(0%)	(0%)	1(100%)	IR
efepime	(0%)	(0%)	(0%)	2(33.3%)	3(75%)	1(50%)	(0%)	(0%)	(0%)	(0%)	1(100%)	(0%)
mpicillin	IR	IR	(0%)	1(16.6%)	1(25%)	(0%)	(0%)	(0%)	IR	(0%)	1(100%)	IR
mpicillin- ılbactum	IR	(0%)	(0%)	1(16.6%)	1(25%)	(0%)	(0%)	(0%)	IR	(0%)	1(100%)	IR
ipieracillin- azobactam	IR	(0%)	(0%)	3(50%)	4(100%)	2(100%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	1(100%)
iprofloxacin	1 (4.34%)	(0%)	(0%)	1(16.6%)	(0%)	(0%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	(0%)
evofloxacin	20(86.95%)	1(12.5%)	(0%)	1(16.6%)	1(25%)	(0%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	(0%)
rimethoprim- ılfamethoxazole	22(95.65%)	3(37.5%)	1(14.28%)	5(83.3%)	(0%)	2(100%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	IR
eftazidime- /ibactam	NA	1(12.5%)	2(28.57%)	4(66.6%)	2(50%)	2(100%)	(0%)	(0%)	1(100%)	(0%)	1(100%)	(0%)
hloramphenicol	23 (100%)	2 (25%)	4(57.14%)	6(100%)	1(25%)	2(100%)	(0%)	(0%)	1(100%)	1(100%)	1(100%)	IR

Figure 3 Proportion of gram-positive organisms (see online version for colours)

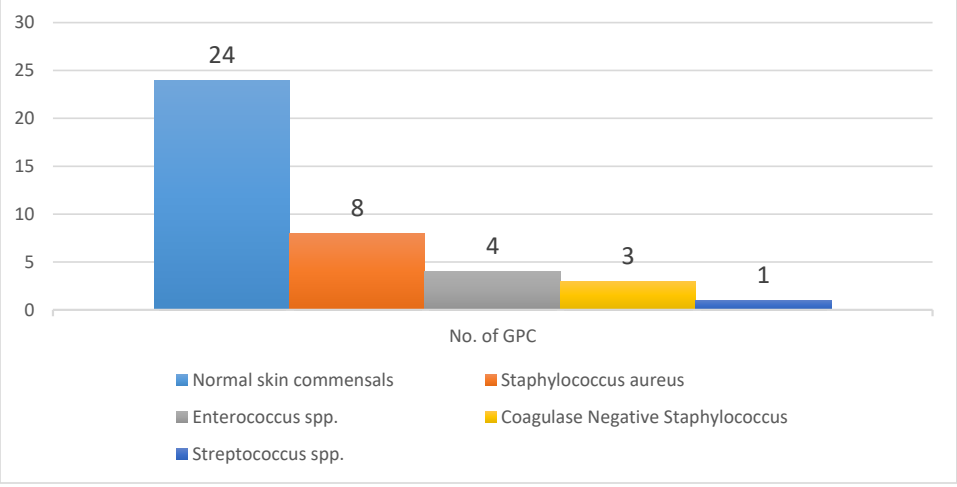


Table 2 Antibiotic susceptibility pattern of GPC

Antibiotics	*Staphylococcus aureus (n = 8) (%)*	*Enterococcus spp. (n = 4) (%)*	*Coagulase negative staphylococci (n = 3)*	*Streptococcus spp. (n = 1) (%)*
Cefoxitin	3 (37.5%)	IR	1 (33.3%)	(0%)
Ampicillin	(0%)	3 (75%)	(0%)	1 (100%)
Penicillin-g	(0%)	3 (75%)	(0%)	1 (100%)
Vancomycin	8 (100%)	4 (100%)	3 (100%)	1 (100%)
Clindamycin	3 (37.5%)	(0%)	(0%)	(0%)
Erythromycin	2 (25%)	(0%)	(0%)	(0%)
Ciprofloxacin	2 (25%)	(0%)	(0%)	(0%)
Moxifloxacin	5 (62.5%)	1 (100%)	3 (100%)	(0%)
Doxycycline	7 (87.5%)	2 (50%)	2 (66.6%)	1 (100%)
Daptomycin	8 (100%)	4 (100%)	3 (100%)	1 (100%)
Trimethoprime – sulfamethoxazole	4 (50%)	IR	(0%)	(0%)
Linezolid	8 (100%)	4 (100%)	3 (100%)	1 (100%)
Rifampin	4 (50%)	(0%)	2 (66.6%)	(0%)

GPC accounted for 37.7% of the isolates including Normal skin flora (60%), Staphylococcus aureus (20%), Enterococcus spp. (10%), Coagulase negative staphylococcus (CONS) (7.5%), Streptococcus spp. (2.5%) (Figure 3). Fungal isolated were candida spp. (7.5%). Gram-negative (54.7%) in our study is more common than gram-positive (37.7%). Gram positive bacteria are more common than gram-negative

bacteria, according to certain research. Mohanty et al. (2017) study shows that gram-positive bacteremia (61.4%) is more than gram negative bacteremia (38.6%). This fluctuation may be a result of seasonal and regional variations. According to reports, temperature is more frequently linked to septicemia, especially in gram-negative bacteria that are clinically significant (Eber et al., 2011).

The most prevalent isolate of gram-negative bacteria in our research, Stenotrophomonas maltophilia, was 100% sensitive to chloramphenicol, 95.65% sensitive to trimethoprim-sulfamethoxazole, and 86.95% susceptible to levofloxacin. A study done by Hamdi et al. (2020) in which quite similar results were observed, sensitivity pattern of stenotrophomonas maltophilia was 98% sensitive to Trimethoprim-sulfamethoxazole, 75% to levofloxacin. The other most common isolate among GNB was Acinetobacter which showed the highest sensitivity 37.5% to the Trimethoprim-sulfamethoxazole followed by Klebsiella which shows 57.14% sensitivity to Chloramphenicol. Amikacin and Chloramphenicol was 100% sensitive to the Escherichia coli. Salmonella typhi shows the 100% sensitivity to Ertapenem, Imipenem, Meropenem and Piperacillin-tazobactam (Table 1).

Gram positive bacteria showed 100% sensitivivity to daptomycin, vancomycin and linezolid. Similar results were observed by Qureshi and Aziz (2012), in which Vancomycin shows 100% sensitivivity. In our study the most common bacteria isolated was, Staphylococcus aureus 100% sensitive to vancomycin, daptomycin and linezolid, 87.5% sensitive to doxycycline, 62.5% to moxifloxacin (Table 2). Tiwari et al. (2013), study shows similar results, in which vancomycin and linezolid 100% sensitive to staphylococcus aureus. Enterococcus spp. showed 100% sensitivity to daptomycin, vancomycin and linezolid, 75% to Ampicillin and Penicillin-G. CONS are also showed 100% sensitivity to daptomycin, vancomycin and linezolid, 66.6% to doxycycline and rifampin. Mohammad et al. (2020). Correlated with our study which shows CONS were 100% sensitive to vancomycin and linezolid.

5 Conclusions

One of the really crucial microbiological techniques is still blood culture. According to the current study, the rate of blood stream infection was 30.1%.The most prevalent isolates from blood cultures were Stenotrophomonas maltophilia and staphylococcus aureus, which is predominant organisms causing septicemia. Gram-negative bacteria are more common than gram-positive bacteria, according to our investigations. The most prevalent strain among the gram-negative bacteria, stenotrophomonas maltophilia, had greater susceptibility to chloramphenicol, trimethoprim-sulfamethoxazole and levofloxacin. The majority of the gram positive bacteria that were identified were Staphylococcus aureus, showed higher sensitivity to daptomycin, vancomycin, linezolid, doxycycline, moxifloxacin.

To help doctors select the best empirical medication, a routine epidemiological analysis of blood culture isolates and evaluation of antibiotic susceptibility is required.

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