International Journal of A J Institute of Medical Sciences 1 (2012) 26-31

Original Research Article

Bloodstream infections caused by Gram-positive cocci: A closer look

Prajna Sharma, Ronald Roche

Department of Microbiology, A.J. Institute of Medical Sciences, Mangalore, India

Abstract

In the last 30 years, the frequency, etiology and epidemiology of blood stream infections (BSIs) have changed with the evolution of medical care, particularly among the increasing number of hospitalized patients who require intensive care. Although gram-negative bacilli were the predominant nosocomial pathogens in the 1970s, Gram-positive cocci have emerged as a more frequent cause of nosocomial BSIs during the 1980s and 1990s. Currently, the 3 most common causes of nosocomial BSIs are coagulase-negative staphylococci, Staphylococcus aureus and enterococci. In our study, 71.42% of BSIs were caused by Gram-positive cocci. Out of which 58.38% was by Staphylococcus aureus and among these 23.4% was methicillin resistant staphylococcus aureus (MRSA). Hence this study stresses the need for the continuous screening and surveillance for antibiotic resistance in hospital settings.

Keywords: Blood stream infections; Bacteraemia; Methicillin resistant staphylococcus aureus; Coagulase negative staphylococci

Received 15th March, 2012; Accepted 2nd April, 2012.

1. Introduction

Bacteraemia is defined as the presence of viable bacteria in the blood and is not necessarily associated with clinical manifestations of the disease. The term bloodstream infection (BSI) has been imposed progressively, and a diagnosis of BSI requires the presence of clinical symptoms of systemic infection in addition to positive blood culture results. BSIs are associated with significant morbidity and mortality, particularly in populations at high risk of infection.[1]

The frequency of these infections, their epidemiology, and the invading organisms have changed in parallel with the evolution of medical care, particularly with the emergence of an increasingly ill and immunocompromised population of hospitalized patients who are often heavily dependent on medical support and indwelling devices. Currently, slightly more than 50% of BSIs are hospital acquired.[2,3] The impact on patient outcome is tremendous; BSI increases the mortality rate, prolongs patient stay in the intensive care unit (ICU) and in the hospital, and generates substantial extra costs.[4] With the development of potent antistaphylococcal β-lactam agents, Staphylococcus aureus gave way to Gram-negative bacilli, often enterobacteriaceae, as the predominant

nosocomial pathogens in the 1970s. Nearly 75% of nosocomial infections were caused by Gram-negative bacilli. From 1981 through 1983, Gram-negative bacilli were recovered in 52% of the episodes of nosocomial BSIs and Gram-positive cocci were recovered in 42%. However, from last decade Gram-positive cocci accounted for 54% of these episodes and Gram-negative rods accounted only for 29%.[5]

It is interesting to study the epidemiology and clinical significance of BSIs caused by Gram-positive cocci. The time of onset of bacteremia varies depending on the infecting organism. Nosocomial bacteremia caused by v. streptococci and s. aureus occurs earlier during hospitalization than does bacteremia caused by Gram-negative bacilli, candida, and enterococci. The average time of onset of bacteremia caused by streptococci and S. aureus is around two weeks after the start of hospitalization. Gram-negative bacilli, candida, and enterococci are encountered in blood cultures on average a week or so after the start of hospitalization. The mean time to onset of bacteremia caused by coagulase-negative staphylococci is around 19 days after the start of hospitalization.[6] Hence, the present study was undertaken in our hospital, to study the bacteriological profile of organisms isolated from blood cultures and their antibiotic sensitivity pattern.

2. Materials and methods

The present study was carried out from January 2010 to December 2010 in the Department of Microbiology, A.J. Institute of Medical sciences, a tertiary care centre in Mangalore, South India.

Blood for culture was collected following strict aseptic precautions. One millilitre (neonates), 3 ml (children) and 5-10ml (adults) blood was collected and inoculated into 10ml of brain heart infusion (1:10 dilution) for neonates and children and 50ml for adults. The culture bottles were incubated at 37ºC aerobically and periodic subcultures were done onto Mac Conkey's agar, blood agar and chocolate agar after 24 hrs, 4 days and 7 days of incubation. The growth obtained was identified by conventional biochemical tests.

The standard disk diffusion test for susceptibility to routine antibiotics was done by modified Kirby-Bauer method. Zone sizes were measured and interpreted according to Clinical and Laboratory Standards Institute's (CLSI) standards.[7] Drug resistant strains in primary screening were further processed for the detection of methicillin resistance and vancomycin resistance in staphylococcus species.

Detection of methicillin resistance was done by cefoxitin disk diffusion method by placing 30 µg Cefoxitin disk on the lawn culture of 0.5 Mc Farland suspension of staphylococcus. After overnight incubation, the zone of inhibition was measured. An inhibition zone of diameter $\leq$ 21mm for S.aureus and $\leq$ 24mm for coagulase negative staphylococci (CONS) indicates methicillin resistance. Quality control strains - methicillin sensitive staphylococcus aureus (MSSA) ATCC 25923 and methicillin resistant staphylococcus aureus (MRSA) ATCC 43300 - were used as negative and positive controls, respectively.[8]

3. Results

During the one year study period, 794 blood culture samples were analyzed. 161 samples showed growth and 633 samples were negative. Gram-positive bacteria were encountered more often (68.93%) than Gram-negative organisms (31.07%). The distribution of species of 161 isolates is reported in Table 1. Antibiotic susceptibility pattern of the s.aureus isolates are shown in Table 2. Among coagulase negative staphylococci (CONS), no methicillin resistance was detected and good antibiotic susceptibility was seen to other drugs mentioned above. Antibiotic susceptibility pattern of v.

streptococci is shown in Table 3. Out of 5 strains of streptococci one was pneumococcus which was uniformly sensitive to all the antibiotics

Table 1: Organisms isolated in blood culture

Organisms isolated	Number	%
Staphylococcus aureus	94	58.38
CONS	12	7.45
Streptococcus	5	3.10
Acinetobacter	18	11.18
Pseudomonas	13	8.07
Klebsiella	7	4.34
E.coli	6	3.72
Candida	7	4.34
Total	161	100

Table 2: Antibiotic susceptibility pattern of staphylococcus aureus

Antibiotics	Sensitive	Resistant
Penicillin (10U)	13	81
Cefazolin (30µg)	81	13
Erythromycin (15µg)	45	49
Clindamycim (2µg)	81	13
Gentamicin (50µg)	91	03
Ciprofloxacin (5µg)	80	14
Cefoxitin (30µg)	72	22
Linezolid (30µg)	94	0
Teicoplanin (30µg)	94	0
Vancomycin (30µg)	94	0
Netilmycin (30µg)	94	0

Table 3: Antibiotic susceptibility pattern of viridans streptococci

Antibiotics	Sensitive	Resistant
Penicillin (10U)	2	2
Ampicillin (10µg)	3	1
Cotrimoxazole (1.25/23.75µg)	3	1
Ciprofloxacin (5µg)	4	0
Gentamicin (50µg)	3	1
Vancomycin (30µg)	4	0

Table 4: Distribution of Gram-positive cocci in our hospital

Organisms isolated		ICU (%)	Wards (%)
S.aureus	MSSA	26 (23.4)	46 (41.44)
	MRSA	6 (5.4)	16 (14.41)
CONS		8 (7.21)	4 (3.60)
Viridans streptococci		1 (0.90)	3 (2.70)
Pneumococci		1 (0.90)	0
Total		42 (37.84)	69 (62.16)

given in Table 3. No enterococcus was isolated in our study.

4. Discussion

Primary bloodstream infection is a leading, preventable infectious complication in critically ill patients and has a negative impact on patients' outcome. Physical signs and symptoms, though useful in identifying possible cases, have limited specificity. Definitive diagnosis is by bacteriologic culture of blood samples to identify organisms and establish antibiotic susceptibility**.**

S. aureus is a major cause of bacteremia, and S. aureus bacteremia is associated with higher morbidity and mortality, compared with bacteremia caused by other pathogens. The burden of s. aureus bacteremia, particularly methicillin-resistant s. aureus bacteremia, in terms of cost and resource use is high. The incidence of s. aureus bacteremia and its complications has increased sharply in recent years because of the increased frequency of invasive procedures, increased numbers of immunocompromised patients, and increased resistance of s. aureus strains to available antibiotics.[9] S. aureus is the second-most common pathogen causing BSIs worldwide, and the leading cause of nosocomial BSIs in Europe.

In the United States, s. aureus is the pathogen that is most frequently isolated from all types of BSI.[1]

In our study percentage of isolation of s. aureus was 58.38% out of which 20.27% was MRSA, which is consistent with the previous studies.[10] Our study isolated 7.45% as CONS from first blood subculture. Although CONS are the most common isolates associated with BSIs in recent years, their role as a cause of morbidity and mortality is difficult to ascertain. Because these organisms commonly contaminate blood cultures, identifying patients with true bacteremia may be difficult.[11]

Organisms causing BSIs vary depending on the location of patients within the institution. CONS are more likely to be isolated from cultures of blood specimens from patients in intensive care settings; whereas v. streptococci and s. aureus are more commonly isolated from ward patients.[6] Enterococcus is isolated with similar frequency from patients in both settings.[12] However, in our study enterococcus was not isolated. Table 4 shows distribution of Gram-positive cocci in our hospital which is consistent with a previous study.[6]

Crude mortality rates among patients with BSIs caused by these Gram-positive cocci range from 17% to 32%; the lowest mortality rates are associated with CONS, and the highest mortality rates have been noted among patients with enterococcal bacteremia.[6,12]

The key treatment options for MSSA bacteremia are the semisynthetic penicillins, cephalosporins and, more recently, the cyclic lipopeptide daptomycin. Current treatment options for MRSA bacteremia include vancomycin, teicoplanin, linezolid, TMP-SMX (trimethoprim -sulphamethoxazole), quinupristin -dalfopristin, and daptomycin. Daptomycin has demonstrated efficacy against both MSSA and MRSA infections and is thus an attractive option for the empirical therapy of suspected s. aureus infection.[13] Vancomycin remains the drug of choice for the Gram-positive bacterial isolates in our setup. MRSA are treated with a combination of ciprofloxacin or vancomycin with amikacin.

5. Conclusion

It seems that the past decade has seen the re-emergence of Gram-positive cocci as the predominant cause of BSIs. In addition, in each of the genera of Gram-positive cocci causing these BSIs, notable antimicrobial resistance is present. It is obvious that options for effective antimicrobial therapy are becoming increasingly limited. Furthermore, in time, the absence of effective therapy will cause morbidity and mortality to increase. Data from the SENTRY Antimicrobial Surveillance Program[14] confirm the importance of Gram-positive cocci as cause of BSIs and the notable antibiotic resistance present in staphylococci, streptococci, and enterococci in both North America and Latin America. Thus, there are mandates to develop new effective antimicrobial therapies for infections caused by Gram-positive cocci, to intensify efforts to limit the spread of resistance in these organisms, and to reduce the nosocomial transmission of infection with these organisms.

References

1. Rio A ,Cervera C, Moreno A, Moreillon P, Miró J. Patients at Risk of Complications of Staphylococcus aureus Bloodstream Infection, Clin Infect Dis 2009;48:S246-S253.
2. Weinstein MP, Towns ML, Quartey SM, Mirrett S, Reimer LG, Parmigiani G, et al. The clinical significance of positive blood cultures in the 1990s: a prospective comprehensive evaluation of the microbiology, epidemiology, and outcome of bacteremia and fungemia in adults. Clin Infect Dis 1997;24:584-602.
3. Bryan CS, Hornung CA, Reynolds KL, Brenner ER. Endemic bacteremia in Columbia, South Carolina. Am J Epidemiol 1986;123:113-127

4. Hugonnet S, Sax H,Eggimann P, Chevrolet JC, Pittet D. Nosocomial bloodstream infection and clinical sepsis. Emerg Infect Diseases 2004;10:76-81.
5. Edmond MB, Wallace SE, McClish DK, Pfaller MA, Jones RN, Wenzel RP. Nosocomial bloodstream infections in United States hospitals: a 3-year analysis. Clin Infect Dis 1999; 29:239-244.
6. Pittet D, Li N, Woolson RF, Wenzel RP. Microbiological factors influencing the outcome of nosocomial bloodstream infections: a 6-year validated, population-based model. Clin Infect Dis 1997;24:1068-1078.
7. Clinical and Laboratory Standards Institute - 2007. Performance standards for antimicrobial susceptibility testing. CLSI approved standard M100-S17;27:32-70.
8. Anand KB, Agrawal P, Kumar S, Kapila K.Comparison of cefoxitin disc diffusion test, oxacillin screen agar, and PCR for mecA gene for detection of MRSA. Ind J of Med Micro 2009;27:27-29.
9. Naber C. Staphylococcus aureus bacteremia: Epidemiology, pathophysiology, and management strategies. Clin Infect Dis 2009; 48:S231-S237.
10. Fluit AC, Jones ME, Schmitz FJ, Acar J, Gupta R, Verhoef J. Antimicrobial Susceptibility and Frequency of Occurrence of Clinical Blood Isolates in Europe from the SENTRY Antimicrobial Surveillance Program, 1997 and 1998. Clin Infect Dis 2000;30:454-460.
11. Karchmer AW, Nosocomial bloodstream infections: organisms, risk factors, and Implications. Clin Inf Dis 2000;31:S139-S143.
12. Maki DG, Agger WA. Enterococcal bacteremia: clinical features, the risk of endocarditis, and management. Medicine (Baltimore) 1988; 67:248-269.
13. Corey JGR, Staphylococcus aureus bloodstream infections: definitions and treatment. Clin Inf Dis 2009;48(4):S254-S259.
14. Pfaller MA, Jones RN, Doern GV, Sader HS, Kugler KC, Beach ML. Survey of blood stream infections attributable to gram-positive cocci: frequency of occurrence and antimicrobial susceptibility and of isolates collected in 1997 in the United States, Canada, and Latin America from the SENTRY Antimicrobial Surveillance Program. SENTRY Participants Group. Diagn Microbiol Infect Dis 1999;33:283-297.

Corresponding author

Dr. Prajna Sharma
Assistant Professor
Department of Microbiology
A.J. Institute of Medical Sciences
Mangalore, Karnataka, India
Mobile: +91 9964571149
Email: drprajnasharma@gmail.com