

Report

Nosocomial blood stream infections in Imam Khomeini Hospital, Urmia, Islamic Republic of Iran, 1999–2001

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حالات العدوى في مجرى الدم، حالات العدوى بمستشفى الإمام الخميني، في أرميا، جمهورية إيران الإسلامية

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الخلاصة: في دراسة استعادية أُجريت على مدى عامين، تمت مراجعة قاعدة معلومات مختبر الميكروبيولوجيا بمستشفى الإمام الخميني في أرميا، بجمهورية إيران الإسلامية، وذلك لتحديد المرضى الذين أصيبوا بتجرثم الدم المستشفوي فيما بين أول أيار/مايو 1999 و31 أيار/مايو 2001، وتحديد العامل المرضي المسؤول ومدى مقاومته للمضادات الحيوية. ومن بين 6492 مريضاً في مختلف أجنحة المستشفى، كان لدى 593 (9.1%) منهم نتائج إيجابية لمزارع الدم التي أُجريت لهم. وكان لدى 85 مريضاً من بين هؤلاء (14.3%) علامات تشير إلى إمكانية وجود تلوث جلدي كامن لديهم. وكانت المكورات الإيجابية الغرام، والتي شملت العنقوديات السلبية المختثرة (الكواغولاز) والعنقودية الذهبية *Staphylococcus aureus* والعقدية الرئوية *Streptococcus pneumoniae* وغير ذلك من المكورات الإيجابية الغرام، كانت مسؤولة عن 42.3% أخرى من الجراثيم المستفردة. وكانت الزائفة الزنجارية *Pseudomonas aeruginosa* هي المستفردة الجرثومية السائدة. وقد تباينت أنماط مقاومة الجراثيم للأدوية تبعاً لنوع الجرثومة، إلا أنها كانت عالية بشكل عام.

ABSTRACT In a 2-year retrospective study, the database of the microbiology laboratory of the Imam Khomeini Hospital was reviewed to identify patients who had nosocomial bacteraemia between 1 May 1999 and 31 May 2001 and identify the pathogen responsible and its resistance to antibiotics. Of 6492 patients in various wards, 593 (9.1%) had positive blood cultures; 85 of those (14.3%) had signs of potential skin contamination. Gram-positive cocci, including coagulase-negative staphylococci, *Staphylococcus aureus*, *Streptococcus pneumoniae* and other Gram-positive cocci, accounted for 42.3% of isolates. Gram-negative bacilli were responsible for another 42.3% of isolates; *Pseudomonas aeruginosa* was the predominant isolate. Patterns of drug resistance varied according to species of bacteria but were generally quite high.

Infections nosocomiales sanguines à l'hôpital Imam Khomeini, Urmia (République islamique d'Iran), 1999-2001

RÉSUMÉ Lors d'une étude rétrospective sur deux ans, la base de données du laboratoire de microbiologie de l'hôpital Imam Khomeini a été examinée pour identifier les patients qui avaient eu une bactériémie nosocomiale entre le 1^{er} mai 1999 et le 31 mai 2001 et déterminer les agents pathogènes responsables et leur résistance aux antibiotiques. Sur 6492 patients hospitalisés dans divers services, 593 (9,1 %) avaient une hémoculture positive ; 85 % de ces derniers (14,3 %) présentaient des signes pouvant indiquer une contamination cutanée. Les cocci à Gram positif, y compris les staphylocoques à coagulase négative, *Staphylococcus aureus*, *Streptococcus pneumoniae* et autres cocci à Gram positif représentaient 42,3 % des isolats. Des bacilles à Gram négatif étaient impliqués dans 42,3 autres pour cent des isolats, *Pseudomonas aeruginosa* étant l'isolat prédominant. La pharmacorésistance variait selon les espèces de bactéries mais elle était généralement assez importante.

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Introduction

Bacteraemia is the presence of bacteria in the bloodstream and may be transient, intermittent or continuous. Bloodstream infections are important causes of morbidity and mortality and many are acquired in hospital. Not only are nosocomial infections increasing, they are more frequently being caused by pathogenic bacteria that are resistant to antibiotics [1–3].

The spectrum of microorganisms that invade the bloodstream has been systematically evaluated in several studies. The organisms most commonly isolated from blood cultures are Gram-positive cocci including coagulase-negative staphylococci, *Staphylococcus aureus*, *Enterococcus* spp. and other organisms likely to inhabit the hospital environment and the gastrointestinal tracts of hospitalized patients [4,5]. Clinical symptoms and signs in patients are not sufficiently reliable to predict bacteraemia with sepsis but early diagnosis and treatment of bloodstream infections are important clinical concerns in order to reduce frequency and increase survival of patients. Epidemiological studies of etiologic agents of bacteraemia and their susceptibility to antimicrobial agents are commonly based on international collaborative studies. However, due to selective pressure from antibiotic use, especially in developing countries, there is a need for local surveillance [6]. Our objective, therefore, was to review the microbiology database of the Imam Khomeini Hospital in Urmia from 1 May 1999–31 May 2001 for cases of bacteraemia.

Methods

Imam Khomeini Hospital is a university affiliated hospital with more than 300 beds. At this hospital blood culture results have

been stored in the central laboratory information system. Each data set includes the following information for each patient: family name, first name, age, ward, sampling date, organism identified and results of antimicrobial susceptibility testing.

As per our standard hospital procedure, nurses disinfect skin with 70% alcohol followed by 2% povidone-iodine before collecting blood samples. Typically 10 mL of blood are collected from adults and approximately 1–5 mL collected from children at each phlebotomy. Blood samples are transferred to blood culture media and immediately transported to the hospital's microbiology laboratory. All blood cultures are incubated at 35 °C for at least 2 weeks. In addition to daily visual examination, blood subcultures are performed 6 to 12 hours after the first incubation on well-mixed media and the inoculum is spread onto chocolate agar, blood agar and eosin methylene blue agar plates. All blood agar and chocolate agar plates are incubated in 5%–10% CO₂ at 35 °C for 48 hours. Negative bottle cultures are re-incubated for an additional 2 weeks.

Isolates are identified by Gram staining and conventional biochemical methods [3,5]. Methods used for identification of Gram positive bacteria are Gram-stain, catalase, coagulase, DNase, bile esculin hydrolysis, growth on NaCl, susceptibility to optochin, colonial morphology and haemolysis. Methods used for identification of Gram-negative rods are catalase, oxidase, indole, motility, H₂S production, fermentation of sugars and other biochemical tests. Susceptibility is tested using the disk diffusion method described by the National Committee for Clinical Laboratory Standards (NCCLS) [7].

A nosocomial bloodstream infection was diagnosed in a clinically ill patient (with fever, chills, hyperventilation and hy-

potension) when 1 or more cultures of blood drawn at least 48 hours after admission yielded a pathogenic organism. If the isolated bacteria was a potential skin contaminant, e.g. *Diphtheroid* spp., *Propionibacterium* spp., *Bacillus* spp., coagulase-negative staphylococci or micrococci, all of the following additional criteria were required for diagnosis of nosocomial bacteraemia: the presence of an intravascular catheter, the initiation of antimicrobial therapy, and either temperature > 38 °C or systolic blood pressure < 90 mmHg. Multiple positive blood cultures yielding the same organism were considered a single infection [4,8–11].

Frequencies were calculated using descriptive statistics and expressed as percentages.

Results

Among the various wards of the Imam Khomeini Hospital, 593 (9.1%) of 6492 patients had positive blood cultures over the 2-year period and were considered to have a nosocomial infection; 85 (14.3%) of these had evidence of potential skin contamination. The predominant contaminant bacteria included *Diphtheroid* spp., *Bacillus* spp., coagulase-negative staphylococci and *Propionibacterium* spp. Of the other bacteria isolated, Gram-positive cocci and Gram-negative bacilli (42.3%) were isolated in equal proportions (42.3% each).

Table 1 shows that coagulase-negative staphylococci (isolated from 111 patients or 18.7%) and *Staph. aureus* (from 107 patients or 18.0%) were the most common causes of nosocomial bacteraemia. *Streptococcus pneumoniae* and other gram-positive cocci were isolated in 4.0% and 1.6% of patients respectively. The most commonly isolated Gram-negative organisms were *Pseudomonas aeruginosa* (from 104

Table 1 Bacteria most commonly isolated from 6492 blood cultures at Imam Khomeini Hospital, 1 May 1999–31 May 2001

Bacteria	No.	%
Total Gram-positive cocci	251	42.3
Coagulase-negative staphylococci	111	18.7
<i>Staphylococcus aureus</i>	107	18.0
<i>Streptococcus pneumoniae</i>	24	4.0
Other gram-positive cocci	9	1.5
Total Gram-negative bacilli	251	42.3
<i>Pseudomonas aeruginosa</i>	104	17.5
<i>Enterobacter</i> spp.	48	8.1
<i>Escherichia coli</i>	41	6.9
<i>Klebsiella pneumoniae</i>	19	3.2
<i>Citrobacter</i> spp.	19	3.2
Other Gram-negative rods	20	3.4
<i>Candida</i> spp.	6	1.0
Contaminants	85	14.3
Total	593	100

patients or 17.5%), *Enterobacter* spp. (from 48 patients or 8.1%) and *Escherichia coli* (from 41 patients or 6.9%) (Table 1). In addition to bacteria, we isolated *Candida albicans* in 5 patients.

The frequency of nosocomial bloodstream infections varied with wards. The highest frequency of positive cultures was in the neonatal ward (for neonates aged under 45 days). In that ward, 323 (54.5%) patients had positive blood cultures. Individual pathogens varied slightly in various wards, e.g. *P. aeruginosa* was most common in the burn (52.8%), haematology (30.5%) and paediatric (18.4%) wards, whereas in the neonatal ward, coagulase-negative staphylococci were predominant (21.9%). The crude mortality rate of bacteraemia due to *P. aeruginosa* was very high; for example, the mortality rates in the burn, haematology and neonatal wards

were 86.6%, 75.0% and 48.7% respectively.

Gram-positive bacteria exhibited a significant rate of antibiotic resistance (Table 2). The antimicrobial resistance rates of *Staph. aureus* were: penicillin 82.6%; oxacillin 40.7%; gentamicin 36.5%; trimethoprim-sulfamethoxazole 7.6% and clindamycin 1.9%. No *Staph. aureus* isolates were resistant to vancomycin or ciprofloxacin. The resistance rates of coagulase-negative staphylococci to the antibiotics were: penicillin 76.8%; oxacillin 61.5%; gentamicin 25.0%; clindamycin 3.2%; and trimethoprim-sulfamethoxazole 6.4%. No coagulase-negative staphylococci were resistant to ciprofloxacin or vancomycin (Table 2).

Antimicrobial resistance levels for the Gram-negative organisms most commonly causing nosocomial blood stream infections were relatively high (Table 3). Resistances rates of *P. aeruginosa* to the antibiotics were: gentamicin 32.2%; tobramycin 21.5%; and amikacin 21.4%. This organism was not resistant to ciprofloxacin. *Enterobacter* spp. resistance rates to the antibiotics were: ampicillin 47.8%; cephalotin 65.2%; ceftizoxim 30.4%; gen-

tamicin 71.7%; tobramycin 56.5%; ciprofloxacin 0%; and trimethoprim-sulfamethoxazole 8.6%. Resistance rates of *E. coli* were: ampicillin 57.4%; cephalotin 62.5%; ceftizoxim 12.5%; gentamicin 22.5%; tobramycin 22.5%; ciprofloxacin 0%; and trimethoprim-sulfamethoxazole 7.5% (Table 3).

Discussion

The rate of contamination of blood cultures in the microbiology laboratory of Imam Khomeini Hospital was high (14.3%), especially in comparison with other hospitals in other countries [6]. One important source of contamination of blood cultures with bacteria such as *Staph. epidermidis*, *Diphtheroid* spp., *Propionibacterium* spp. and *Bacillus* spp. is the insufficient disinfecting of skin during blood sampling. Further complicating matters, in our hospital physicians often submit 1 instead of 3 samples, making the interpretation of blood culture results difficult [9,12,13].

The spectrum of microorganisms that invade the bloodstream has been systematically evaluated in several studies. In a surveillance study of nosocomial bloodstream

Table 2 Antimicrobial resistance of Gram-positive cocci isolated from blood cultures of 251 patients at Imam Khomeini Hospital, 1 May 1999–31 May 2001

Antibiotic	<i>Staphylococcus aureus</i> (%)	Coagulase-negative staphylococci (%)	<i>Streptococcus pneumoniae</i> (%)
Penicillin	82.6	76.8	31.8
Oxacillin	40.7	61.5	22.7
Vancomycin	0	0	0
Gentamicin	36.5	25.0	Not tested
Ciprofloxacin	0	0	0
Clindamycin	1.9	3.2	0
Trimethoprim-sulfamethoxazole	7.6	6.4	0

Table 3 Antimicrobial resistance of Gram-negative bacilli isolated from blood cultures of 251 patients at Imam Khomeini Hospital, 1 May 1999–31 May 2001

Antibiotic	<i>Pseudomonas aeruginosa</i> (%)	<i>Enterobacter</i> spp. (%)	<i>Klebsiella pneumoniae</i> (%)	<i>Escherichia coli</i> (%)
Ampicillin	(not tested)	47.8	72.2	57.4
Cephalotin	(not tested)	65.2	27.3	62.5
Ceftizoxim	72.3	30.4	16.6	12.5
Gentamicin	32.2	71.7	27.7	22.5
Tobramycin	21.5	56.5	11.1	22.5
Amikacin	21.4	2.1	0	0
Ciprofloxacin	0	0	15.5	0
Trimethoprim–sulfamethoxazole	90.1	8.6	16.6	7.5

infection at 49 hospitals in the United States of America over a 3-year period, over 10 000 bloodstream infections were detected [8]. Gram-positive organisms were responsible for 64% and Gram-negative organisms were responsible for 27%; 8% were caused by fungi [8]. In our study, 42.3% of infections were caused by Gram-positive cocci and 42.3% by Gram-negative bacilli. The rate of isolation of fungi (1.0%) in our study was low in comparison with other studies [6].

The role of coagulase-negative staphylococci in bacteraemia continues to be controversial. Until the 1970s, coagulase-negative staphylococci were mainly recognized as a contaminant, i.e. as part of the skin flora. Since then, several studies have reported increasing incidence of infections due to coagulase-negative staphylococci [14,15]. In one study, coagulase-negative staphylococci and *Staph. aureus* were the first and second most common etiologic agents respectively causing nosocomial bloodstream infections in the USA [16]. In another study, *Staph. aureus* and *E.coli*

were the most commonly isolated bloodstream infections, followed by coagulase-negative staphylococci and enterococci [17]. *Klebsiella* spp., *Enterobacter* spp., *P. aeruginosa*, *Strep. pneumoniae* and β -haemolytic streptococci were also among the 10 most frequently reported species in both the USA and Canada [17].

In our study coagulase-negative staphylococci and *Staph. aureus* were isolated in almost half the total cases of bacteraemia. Other recent studies report similar results [7,18]. In a general hospital in Kerman in the southern part of our country, *Staph. aureus* and coagulase-negative staphylococci were isolated from 179 blood specimens (45.7%) [18]. This is similar to our study (36.8% for all specimens). *Strep. pneumoniae* (4.0%) was isolated from blood cultures of 24 patients. This organism is a major and well-known cause of community-acquired infections, but there is increasing interest in its role in the epidemiology of hospital-acquired infection [19].

The resistance of *Staph. aureus* and coagulase-negative staphylococci to antibiotics such as penicillin and oxacillin was high. Nearly 50% of isolates were resistant to oxacillin. Fortunately we did not observe any resistance of *Staph. aureus* and coagulase-negative staphylococci to vancomycin.

Gram-negative bacilli accounted for approximately half of the nosocomial bloodstream infections and their resistance to commonly used antibiotics was also relatively high. Nosocomial bloodstream infections caused by *P. aeruginosa* were prevalent in the burn ward. The high mortality rate from septicemia caused by *P. aeruginosa* may be due to resistance of this organism to numerous drugs and also to

the poor management of patients [4,5] by physicians.

Conclusion

Our study provides data on the rate of nosocomial infection in our hospital and the organisms involved as well as the antibiotic resistance. This could be of value in the proper antimicrobial therapy of bacteremia in our hospital. In addition, this ongoing surveillance project will continue to track nosocomial bloodstream infections in Imam Khomeini Hospital and emerging trends in antibiotic resistance. Plans should be made to evaluate the relationship between antibiotic use in the hospital and patterns of antimicrobial resistance.

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