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Full Length Article

Source, pattern and antibiotic resistance of blood stream infections in hematopoietic stem cell transplant recipients

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KEYWORDS

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Abstract Mucositis developing as a result of myelo-ablative high dose therapy administered prior to hematopoietic stem cell transplantation (HSCT) is associated with the risk of bacteremia. The aim of the present study was to detect the pattern of bacteremia coinciding with the present practice of HSCT, to study the contribution of health-care associated infection (HAI) to the pattern of infection, in the context of the problem of antibiotic resistance in HSCT recipients.

Patients and methods: This is a retrospective, single center study including patients who developed febrile neutropenia (FN) among HSCT recipients in one year duration.

Results: Ninety FN episodes were recorded in 50 patients. Out of 39 positive blood cultures, Gram negative rods (GNR) were the predominant pathogens, constituting 67% ($n = 26$) of isolated organisms, while 33% of infections were caused by gram positive cocci (GPC) ($n = 13$). Bacteremia was significantly associated with central venous line (CVL) infections and gastroenteritis (diarrhea and vomiting) with a p-value 0.024, 0.20 and 0.0001, respectively. Multi-drug resistant organisms (MDROs) were identified in 27 (69%) of the 39 positive blood cultures.

Conclusion: In one year duration, gram negative pathogens were the predominant causes of infection in HSCT recipients with high rates of MDROs in our institution. Gastroenteritis and central venous line infections are the main sources of bacteremia.

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Introduction

Despite the advances in conditioning, supportive care and prophylaxis, infection remains a major cause of morbidity and mortality in HSCT due to continued development of antimicrobial resistance and the emergence of new pathogens [1].

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On investigating normal microflora and their role in maintaining the micro biome it was found that they may prevent colonization of pathogenic microorganisms including MDROs. This is achieved by many ways either competition of space and resources or a complex immunologic and biochemical interactions. The overuse and empirical introduction of antibacterial agents may play a role in the appearance of MDROs. The new approach now is to maintain an intact micro biome through the usage of micro biome-sparing antimicrobial therapy and discovering ways to augment host protective mechanisms thus maintaining a healthy micro biota [2].

The most predominant type of bacterial infection when patients are neutropenic is bacteremia. The main sources of bacterial infection include central venous cathetes, oral flora and gut flora via bacterial translocations [3]. Health-care associated infections (HAI) present evolving challenges to the successful management of infectious complications in this expanding patient population. Therefore, the aim of the present study was primarily to detect the pattern of bacteremia in HSCT patients coinciding with the present practice of HSCT, to study the contribution of HAI to the pattern of infection, and, to investigate the problem of antibiotic resistance on the course and outcome in transplant patients in our institution in one year duration.

Patients and methods

This is a retrospective, single center study conducted at the Microbiology laboratory of the clinical pathology department in collaboration with the Bone Marrow Transplant unit, at The National Cancer Institute of Cairo (NCI), Egypt, in one year duration. During the period extending from the first of January till the end of December 2009, 80 patients had undergone HSCT for different hematological malignant disorders. The study was approved by Ethics Committee of NCI and an informed consent was obtained from all participants.

During this period, 90 bacteremic episodes were recorded in 50 patients only who showed neutropenia and fever. Their data included were age, diagnosis, state of disease, clinically documented infection (CDI) especially central venous line (CVL) and lower respiratory tract infections (LRTI), duration of episode, and broad spectrum antibiotics given with the need of shift to second line. All patients were hospitalized during the study period and if a patient developed fever >48 h after hospitalization the case was considered febrile neutropenia.

Definitions

In the context of HSCT, fever was defined as a single oral temperature of 38.3 °C, or a temperature more or equal to 38.0 °C for 1 h at 2 times with a minimum interval of 12 h. Neutropenia described an absolute neutrophil count (ANC) $<0.5\times10^9$/L or $<1.0\times10^9$/L, with decline predicted over the next two days [4]. Lower respiratory tract infections were defined as any new infiltrate arising within 48 h before or after the onset of fever and neutropenia.

Microbiology

Two blood culture sets were usually drawn from each patient within the first day of fever from two separate veins. If the cannula site, portacath, or central venous catheter (CVC) was suspected as the source of infection, a blood sample was obtained from it as well. Collected blood was directly injected into Bactec® (Becton Dickinsion, USA) culture vials and they were incubated in the Bactec 9050® incubator. When the blood culture revealed a potential contaminant, another positive blood culture, or other clinical evidence of ongoing blood stream infections (BSI), such as rigors, hypotension, or documented infection at a second site with the same organism, was required for confirmation of true infection. Identification of isolates was carried out utilizing MicroScan® dried gram negative MIC/Combo and dried gram positive MIC/Combo panels Siemens Healthcare Diagnostics Ltd. (Sir William Siemens Sq. Frimley, Camberley, UK GU16 8QD) for gram-negative and gram positive organisms. The panel of antibiotics used for gram negative included: amikacin, amoxicillin-clavulanate, ampicillin-sulbactam, cefepime, norofloxacin, ceftazidime, cefotaxime, ciprofloxacin, levofloxacin, gentamicin, tobramycin, imipenem and meropenem. For gram positives the following panel was used amoxicillin-clavulanate, oxacillin, cefepime, cefotaxime, ciprofloxacin, levofloxacin, clindamycin, imipenem, linezolid and vancomycin. Antimicrobial susceptibility testing both manual using antibiotic disk on Muller Hinton agar using petri dishes at 37 °C for 24–48 h. [5], and automated testing using minimal inhibitory concentration (MIC) by the Microscan was determined by using the criteria established by the National Committee for clinical laboratory standards (CLSI) [6]. The multidrug resistant organism phenotype was defined as diminished susceptibility to $\geqslant3$ antibiotics of the antibiotic groups [7].

Management

According to NCI guidelines, empirical treatment was given to F&N patients after withdrawal of blood samples for blood cultures. The first line of empirical regimen included a third generation cephalosporins and amikacin double agent antimicrobial therapy for 3 days, antibiotics were continued if the patient showed response. If fever persisted, treatment was given according to microbiological results and/or clinical examination necessitating shift or addition of antibiotics; whereas if culture results were not indicative, a carbapenem and antifungal were given. Both antibiotics were continued until the patient became afebrile and ANC exceeded 0.5×10^9/L. After recovery, the patients with persistent fever or clinical symptoms related to the infectious episode were placed under close follow up for 2 weeks for verification.

Statistical methods

Data were analyzed using SPSSwin statistical package version 17. Chi-square test or Fisher's Exact test was used to test the relation between qualitative variables. A p-value less than 0.05 was considered significant. All tests were two tailed.

Results

Patient characteristics

In the period from January first to the end of December 2009, blood stream infections were detected in 39 of 90 (43%) febrile

neutropenic episodes identified in 50 HSCT patients. The patients' age ranged from 3 to 62 years, with a mean age of 29 ± 15 years. There were 28 (56%) males and 22 (44%) were females. The patients' diagnoses were AML, ALL, NHL, and other malignancies (CML, HD, MDS, and NB) in 24 (48%), 12 (24%), 7 (14%), and 7 (14%) patients; respectively.

Clinical features

Central venous line infection, diarrhea and LRTI were recorded in 16 (18%), 15 (17%) and 10 (11%) episodes; respectively.

Microbiology

Out of 39 positive blood cultures, Gram negative rods (GNR) were the predominant causative agents of BSI, constituting 67% (n = 26) of isolated organisms, while 33% of infections were caused by gram positive cocci (GPC) (n = 13). Of the GPC isolated, 90% were oxacillin resistant. Among the GNR, *Enterobacteriaceae* were isolated from 15 cases (58%) and, non-fermenters from 11 including the mixed samples (42%). Bacterial growth was detected in 16 (17.8%) of CVL blood samples, GNR were identified in 10 CVL infections and GPC in 6. The organisms isolated from blood cultures are summarized in Table 1. Table 2 demonstrates the different clinical findings in relation to presence or absence of bacteremia. Multi-drug resistance was identified in 27 (69%) of the 39 positive blood cultures. Fig. 1 illustrates the results of antibiotic sensitivity of isolated organisms causing BSI.

Table 1 Isolated organisms from 39 bacteremic episodes in stem cell transplant recipients with febrile neutropenia.

Organisms	Number (%)
Gram positive organisms	13(33)
Coagulase negative staphylococci	09(23)
Streptococcus spp.	02(05)
Stapylococcus aureus, MRSA	02(05)
Gram negative organisms	26(67)
Non-fermenters	11
Acinetobacter sp.	05(13)
Pseudomonas sp.	05(13)
Stenotrophomonas maltophilia	01(02)
Enterobacteriaceae	15
E. coli	01(03)
Klebsiella spp.	03(08)
Enterobacter spp.	08(21)
Citrobacter spp.	02(05)
Serratia spp.	01(02)
Mixed gram positive and gram negative (n = 3); included in GNR	

MRSA = Methicillin resistant *Staphylococcus aureus*.

Health-care associated infections

An outbreak was detected at the Bone Marrow Transplant Unit (BMT) in NCI by the Microbiology laboratory staff

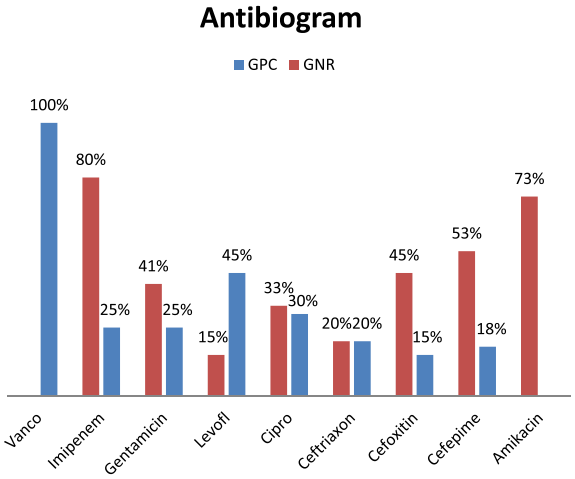

Fig. 1 Antibiotic sensitivity of Gram negative (GNR) and gram positive organisms (GPC) isolated from positive blood cultures of febrile neutropenicstem cell transplant recipients in one year duration.

Table 2 Clinical characteristics of febrile neutropenic stem cell transplant recipients in relation to results of blood culture in 90 episodes.

Factors	Blood culture no growth n = 51 N (%)	Result bacterial growth n = 39 N (%)	P-Value
Central venous line infection, n = 16			
No infection, n = 74	5 (9.8)	11 (28.2)	0.024
	46 (90.2)	28 (71.8)	
Lower respiratory tract infection, n = 10			
No infection, n = 80	5 (9.8)	5 (12.8)	0.741
	46 (90.2)	34 (87.2)	
Diarrhea, n = 15	4 (7.8)	11 (28.2)	0.020
No diarrhea, n = 75	47 (92.2)	28 (71.8)	
Vomiting, n = 8	0 (0.0)	8 (20.5)	0.001
No vomiting, n = 82	51 (100.0)	31 (79.5)	
Duration of fever, < 7	35 (68.6)	21 (53.8)	0.152
⩾7	16 (31.4)	18 (46.2)	
Need of antibiotic shift, n = 26			
No shift, n = 64	7 (13.7)	19 (48.7)	< 0.001
	44 (86.3)	20 (61.3)	

and *Enterobacter cloacae* was found to be the major cause during the month of August; 2009. The outbreak constituted of 7 patients who were diagnosed during an episode of neutropenia fever with *E. cloacae* isolated from their blood cultures. Investigations were carried out by the infection control team to detect the cause of the outbreak and to prevent the spread of infection. The cause of this outbreak was due to the presence of new nurses at the BMT unit; who were not familiar with the strict aseptic procedures, as they had not received appropriate training before working at BMT unit. These nurses were dismissed from work at BMT. The 7 patients were properly treated and came out alive. No further cases of *E. cloacae* infection occurred afterward.

Course and outcome

The median duration of neutropenic fever episodes was 5 days. The first line of empirical antibiotic therapy (cefepime and amikacin) was used in 43 of the 90 episodes; second line (carbapenem/amikacin) was given in 36 episodes, whereas monotherapy (cefepime or amikacin) was given in 11 episodes. Antibiotic shift was required in 26 (28.9%) episodes, while vancomycin was added in 30 (33.3%) episodes. Crude mortality was 10% (5/50), with 60% infection attributed mortality.

Discussion

Despite the advances in conditioning regimens, supportive care and prophylaxis, infection remains a major cause of morbidity and mortality in HSCT patients. In the present study, blood stream infections were detected in 39 of 90 (43%) febrile neutropenic episodes identified in 50 HSCT patients. Gram negative organisms were the predominant bacterial pathogens, accounting for 67% of bacterial isolates. Similar results were previously reported in HSCT recipients. In a retrospective study investigating the impact of BSI on outcome of 246 allogeneic HSCT recipients between 1999 and 2006, GNR constituted 54% of the bacterial isolates [8]. In another study, it was found that gram positive cocci accounted for 60% of bacterial isolates as causes of BSI in allogeneic HSCT patients who received levofloxacin prophylaxis [9]. In the present study, GNR isolates were significantly associated with prolonged febrile episodes and requirement of change of empirical line of therapy. Antibiotic shift was needed in 26 episodes (29%), of which 54% were for GNR pathogens. The shift was due to the fact that the empirical guideline in BMT unit at NCI includes antibiotics which unfortunately does not cover extended spectrum β-lactamase (ESBL) and a shift was needed whenever an ESBL Gram negative organism was detected. Similarly, Liu and his colleagues (2011) reported that HSCT patients with GNR pathogens as causes of BSI had a significantly increased length of hospital stay ($p = 0.014$), and a significantly greater 6-month mortality ($p = 0.021$) [8].

In the current study, the CVCs and GIT were the main portals of entry of bacteria, especially GNR isolates. Central venous line infection, and diarrhea were coinciding with BSI in 16 (18%), and 15 (17%) episodes; respectively; and were significantly associated with GNR isolates. Health care associated infections represent another important source of infection in these vulnerable patients. In the current study, an outbreak was recorded during the month of August, coinciding with the new nurses in the unit who lacked the knowledge of proper infection control measures necessary to prevent cross infection.

In our group of HSCT recipients, a high frequency of multidrug resistance was encountered in bacterial pathogens isolated from blood cultures. Multi-drug resistance was identified in 69% (27/39) of bacteria isolated from positive blood cultures. A high incidence of MDROs causing blood stream infections was recently reported to be an increasing problem in critically ill patients as a result of previous use of antibiotics and transfer from other hospitals [10]. *Escherichia coli* resistance to quinolones has increased from 13% to 46% in a recent report from a single cancer center, as a consequence of widespread use of quinolones as prophylaxis for neutropenic patients; and 18% of *E. coli* recovered from BSI were ESBL producing [11]. In patients receiving renal transplantation, infections caused by antibiotic resistant pathogens conferred a worse prognosis [12]. In HSCT recipient's previous intake of antibiotics was significantly associated with the development of antibiotic resistance [8,9]. Levofloxacin with penicillin or doxacycline prophylaxis also contributes to the emergence of resistant GN infections in allogeneic HSCT recipients over time [13].

The problem of antimicrobial resistance is highlighted by The World Health Organization (WHO) and combating antimicrobial resistance has been selected as the theme for World Health Day 2011 [14]. The fear of carbapenemase producing acquisition by GN pathogens is more aggravated in cancer patients especially HSCT recipients [15]. As the prevalence rates of resistance in GN pathogens increases, the treatment options available become limited. Thus, the WHO is calling all parties involved in this problem to act and take responsibility for combating antimicrobial resistance [14].

In agreement with our findings it is stated that in National Comprehensive Cancer Network (NCCN) that vascular access infections are common as a consequence of the utility of vascular associated device in patients undergoing intensive or cyclic chemotherapy [16]. The gut plays a prominent role in the development of resistant micro-organisms in the community, long term care facilities and hospitals [17]. The emergence of MDROs in the gut maybe related to the ingestion of highly pathogenic microorganism or to antibiotic induced alterations of the gut micro biome [2]. This was aided by the finding that bacteremia was significantly associated with GIT infections in our patients.

Thus, we conclude that Gram negative pathogens are the predominant organisms isolated from blood cultures of transplanted patients at our institute. Although coagulase negative Staphylococci are considered normal flora, but in the presence of a central line and infection with gram negative organisms in association of febrile neutropenia, we consider these patients to be suffering from mixed infections. Endogenous bacteria through GIT constitute an important source of infection in HSCT recipients. Gastroenteritis and central venous line infections are the main sources of bacteremia. Health care acquired infections constitute an important source of infection in BMT unit at NCI. Due to the presence of newly appointed nurses in the wards a major outbreak was recorded during this period and attributed in its major part to their lack of proper hygiene and the adequate use of safety measures to prevent the spread of infections. Thus, extreme measures should be taken in order to prevent the occurrence of outbreaks through careful monitoring and continuous education of healthcare workers.

The high rates of antibiotic resistance encountered in HSCT patients at our institute are alarming and necessitate every effort to control this problem. As health care acquired infections caused by multi-drug resistant organisms complicate therapy and limit treatment options, efforts should be directed toward continuous monitoring to ensure strict adherence to infection control measures and proper use of antibiotics to minimize the incidence of the aggravating problem of MDROs.

Conflict of interest

None declared.

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