An eight-year review of blood culture and susceptibility among sepsis cases in an emergency department in North-eastern Malaysia

Hashairi, F.[1*], Hasan, H.[2], Azlan, K.[1] and Deris, Z.Z.[3]
[1]Department of Emergency Medicine, Hospital Universiti Sains Malaysia 16150 Kubang Kerian, Kelantan, Malaysia
[2]Infection Control and Hospital Epidemiology Unit, Hospital Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia
[3]Department of Medical Microbiology and Parasitology, School of Medical Sciences, Universiti Sains Malaysia Health Campus 16150 Kubang Kerian, Kelantan, Malaysia
[*]Corresponding author email: hashairi@kk.usm.my
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Abstract. An understanding of common pathogens and their antibiotic sensitivity patterns is critical for proper management of sepsis in Emergency Department (ED). The goal of the study was to identify common organisms isolated from blood cultures of patients attended to ED and their antimicrobial susceptibility. Beginning from 2002, all cases of positive blood culture collected by the ED, Hospital Universiti Sains Malaysia (HUSM) were recorded and analysed. Over the period of eight years, we documented 995 cases of positive blood cultures. Of these samples, 549 (55.2%) were Gram-negative bacteria; 419 (42.1%) were Gram-positive bacteria; 10 (1.0%) were anaerobic organisms; 10 (1.0%) were fungus; and 7 (0.7%) cases were mixed organisms. Gram-negative bacteria were observed to develop more resistance to antimicrobial agents, especially those commonly used in an outpatient setting with less than 80% sensitivity to ampicillin, cotrimoxazole and ciprofloxacin. By contrast, there has been no marked change in the sensitivity trends of Gram-positive bacteria over the same period. In conclusion, ED physicians are more equipped to initiate empirical antimicrobial therapy especially when dealing with possibility of Gram-negative sepsis.

INTRODUCTION

Sepsis is becoming a major concern in Emergency Departments (EDs), and patients with even mild symptoms are often admitted to the ED (Martin, 2008). An estimated 0.40% of ED admissions in the United States are related to sepsis (Strehlow *et al.*, 2006), and approximately 61% of sepsis cases first present to the ED (Talan *et al.*, 2008). Gram-negative sepsis, in particular, is associated with a high rate of mortality. As a result, emergency physicians must quickly identify at-risk patients and aggressively implement an appropriate antibiotic regimen (Scheineder, 2004).

Identification of the organisms and antimicrobial susceptibilities can be important in subsequent management; however, definitive identification of the pathogen responsible for septic shock is difficult during ED evaluation. Testing of blood cultures is useful for confirming the presence of a pathogen in the blood of a patient, but this method suffers from low sensitivity, which limits its diagnostic utility (Ramos *et al.*, 2004). While obtaining the necessary cultures prior to antimicrobial treatment is critical for pathogen identification and evaluation of its sensitivity to antibiotics, this practice should not delay therapy unreasonably (Talan *et al.*, 2008). ED

physicians must recognise and distinguish minor infections from acute, life-threatening infections (Martin, 2008).

The aim of this study was to identify the common blood isolates and their antimicrobial susceptibility patterns in sepsis patients presented to ED and the results obtained could be used to guide the emergency physician on the treatment to be given to the patients while waiting for the definitive pathogen identification.

MATERIALS AND METHODS

Study design

Beginning in 2002, all cases of positive blood culture from the ED of HUSM were recorded using WHONET 5.2 software (World Health Organization; http://www.who.int/drug resistance/whonetsoftware/en/). A retrospective study was conducted by retrieving all data from blood culture specimens collected by the ED between 1 January 2002 and 31 December 2009. If an organism was repeatedly isolated from any particular patient, only the first isolate was included in the analysis.

Study setting

HUSM is an 800-bed, tertiary teaching hospital located in North-eastern Malaysia. Its ED manages approximately 4,500-5,000 cases per month, and all patients entering the ED undergo triage. Patient's that fulfilled criteria for sepsis according to International Sepsis Guidelines (Dellinger *et al.*, 2008) and required intravenous antibiotics were taken for blood cultures before starting the treatment. This hospital is supported by a 24-hour Medical Microbiology and Parasitology Laboratory, which is able to process up to 20,000 blood cultures each year.

Procedure for the collection of blood culture in the ED

Blood was drawn under aseptic conditions. The puncture site was cleansed with a chlorhexidine solution, and the surrounding skin was draped before venepuncture. At least 5 ml of blood was inoculated into each aerobic or anaerobic culture bottle. One or two sets of blood cultures were collected from each patient before beginning antibiotic therapy. The specimens were sent immediately to the microbiology laboratory for processing.

Culture conditions and bacterial identification

Each blood culture was processed according to the recommendations of the Clinical Laboratory Standard Institute (CLSI). In 2002, before the introduction of BACTEC™ series (Becton Dickinson Microbiology Systems, Sparks, Md., USA), the transport medium used was liquid broth for aerobic cultures and thioglycolate broth for anaerobic cultures. The inoculated bottles were incubated at 37ºC, and turbidity was assessed daily by visual inspection. Blind subcultures were performed every other day whenever turbidity was not observed. After the implementation of the semi-automated BACTEC™ 9000 series (Becton Dickinson Microbiology Systems, Sparks, Md., USA) system in 2003, the practice of blind subcultures was discontinued. All isolates were tested for antimicrobial susceptibility. Blood cultures were incubated for at least five days before being discarded as negative.

Antimicrobial susceptibility testing

Antimicrobial susceptibility testing was conducted using the Kirby-Bauer disc diffusion method, and the breakpoint of the zone of inhibition for each sample was interpreted according to CLSI. Based on the results of the antimicrobial susceptibility testing, each isolate was classified as either sensitive or resistant. To guide the appropriate choice of antimicrobial therapy by physician, organisms displaying an intermediate-resistant phenotype were reported as resistant. The antibiotic discs utilised in this study were ampicillin, gentamicin, ceftriaxone, cefuroxime, ceftazidime, cotrimoxazole or imipenem for Gram-negative isolates and for Gram-positive isolates, ampicillin, gentamicin, cotrimoxazole, ciprofloxacin, oxacillin (substituted with cefoxitin after 2007), erythromycin, and penicillin were used.

RESULTS

Over an eight-year period, an overall positivity rate of 16.7% was reported for the 5,957 blood cultures collected in the ED. Of the 995 organisms that were isolated, 549 (55.2%) were Gram-negative bacteria; 419 (42.1%) were Gram-positive bacteria; 10 (1.0%) were anaerobic organisms; 10 (1.0%) were fungal species; and 7 (0.7%) were mixed-species isolates. The total number of blood isolates collected annually in the ED over the eight-year study is shown in Figure 1. The yearly positivity rates fluctuated between 9.5-40.8%. Isolates containing coagulase-negative staphylococci, Gram-positive rods and mixed-species cultures were assumed to be contaminated; these contaminants were responsible for 2.7-14.3% of positive cultures.

A Gram-negative bacterium, *Salmonella* Typhi, was the most commonly isolated microorganism (331 total cases); in fact, a peak in the number of Gram-negative isolates collected coincided with an outbreak of typhoid in the region during 2005 (174 reported cases). The two other Gram-negative bacteria frequently isolated between 2002 and 2009 were *Escherichia coli* (71 cases) and *Klebsiella pneumoniae* (48 cases). Ten isolates of *Burkholderia pseudomallei* were also recovered during this study.

The major Gram-positive bacteria isolated from blood cultures were coagulase-negative staphylococci (220 cases), *Staphylococcus aureus* (89 cases), *Streptococcus pneumoniae* (13 cases), beta-haemolytic, Group A streptococci (8 cases) and other streptococci/enterococci (44 cases). None was the cause of any suspected outbreak.

The Gram-negative bacteria were observed to gradually develop greater resistance to all antimicrobial agents regularly prescribed in outpatient settings. For example, the percentage of Gram-negative isolates sensitive to ampicillin was reduced from 85.5% in 2002 to 54.3% in 2009. Similar trends were detected for other antibiotics, such as cotrimoxazole and ciprofloxacin, but not to the degree observed with ampicillin (Figure 2). Interestingly, there was a temporary reversal in the sensitivity pattern of *Salmonella enterica* serovar Typhi (*Salmonella* Typhi) in 2005 due to an outbreak of ampicillin-susceptible strains. There was no obvious change in sensitivity of Gram-negative bacteria to any of the other antimicrobials. Similar observations were
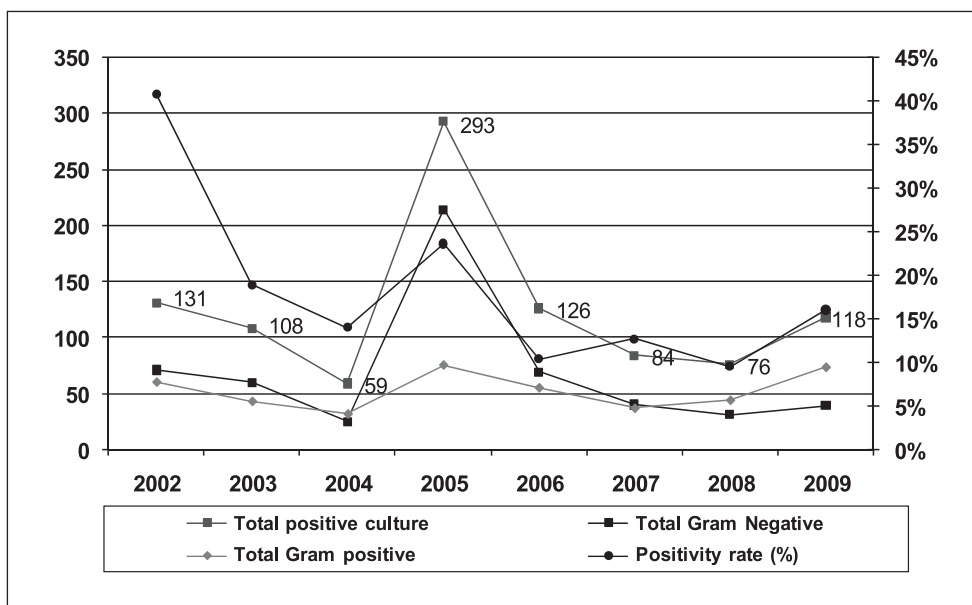

Figure 1. An eight-year trends of total blood culture positive and positivity rate from Emergency Department, Hospital Universiti Sains Malaysia
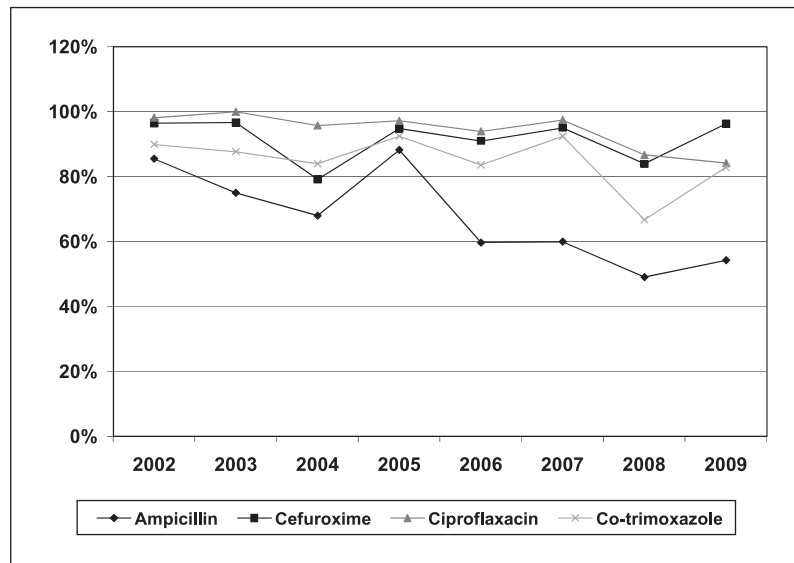
Figure 2. The sensitivity of commonly used oral preparation antimicrobials to Gram negative blood isolates from Emergency Department, Hospital Universiti Sains Malaysia

Table 1. An eight-year trends of sensitivity patterns of bacteria blood isolates from Emergency Department, Hospital Universiti Sains Malaysia. The susceptibility data of Gram negative isolates to commonly used oral antimicrobials were not included in the table. NT –Not tested

Antimicrobial Agents	Bacteria group	Percentage of susceptible							
		2002	2003	2004	2005	2006	2007	2008	2009
Penicillin	Gram positive	46.6	48.8	39.4	38.2	42.6	54.1	42.7	27.4
Ampicillin	Streptococci	100.0	100.0	100.0	66.7	76.9	90.0	100.0	83.3
Ciprofloxacin	Gram positive	NT	NT	NT	NT	97.5	100.0	89.7	94.4
Co-trimoxazole	Gram positive	89.6	87.5	95.5	89.1	85.7	86.7	86.2	75.9
Gentamicin	Gram negative	95.7	96.2	83.3	96.2	94.1	95	85.2	96.6
Gentamicin	Gram positive	78.4	93.3	95.7	89.5	90.5	88.0	87.5	87.0
Ceftriaxone	Gram Negative	100.0	100.0	95.8	99.5	95.5	95	83.9	95.7
Ceftazidime	Gram Negative	100.0	100.0	91.7	98.6	97.1	97.5	90.0	96.7
Imipenem	Gram negative	100.0	100.0	100.0	99.0	98.5	97.5	100.0	100.0
Erythromycin	Gram positive	76.7	78	90.3	89.5	81.5	86.5	81.0	68.3
Oxacillin	Staphylococci	82.4	68.8	95.5	87.1	87.8	87.2	90.0	81.5

documented for Gram-positive bacteria over the past eight years (Table 1).

DISCUSSION

Gram-negative bacilli were the predominant pathogens in our ED setting and accounted for more than half of blood isolates. During infection, Gram-negative bacteria are known to release an outer membrane component, lipopolysaccharide. This molecule stimulates production of cytokines, which can lead to tachycardia and hypotension (Scheineder, 2004). Despite the use of antimicrobial therapy and a recent understanding of the mechanisms of sepsis, the mortality rate from Gram-negative sepsis remains high.

Early treatment of sepsis, particularly when due to Gram-negative bacteria, is

crucial to save a patient's life. Implementation of early goal-directed therapy (EGDT) in the treatment of sepsis has been shown to reduce morbidity and mortality. In addition to the practices of fluid resuscitation and maintenance of adequate oxygenation, which are both involved in EGDT protocol, empirical therapy using the proper antibiotic regimen can also lower the risk of mortality. On the other hand, as a recent report by (Kumar *et al.*, 2009) revealed, early administration of an inappropriate antimicrobial may lead to as much as a five-fold reduction in survival rate for patients with septic shock.

Selection of an initial antimicrobial therapy should be based on local epidemiology and resistance patterns of pathogens. Our results showed that Gram-negative bacteria developed greater resistance to antimicrobial agents commonly used in an ED setting. *In vitro* susceptibility to ampicillin, for example, decreased to less than 80%. Therefore, ampicillin is likely not a suitable antibiotic for empirical therapy when a community-acquired, Gram-negative infection is suspected. Two other antimicrobial agents utilised regularly in an outpatient setting, cotrimoxazole and ciprofloxacin, exhibited a similarly reduced effectiveness; nonetheless, data collected in 2009 indicated that the susceptibility rate of Gram-negative infections to these two agents remained above 80%. The increasing trend in antimicrobial resistance is possibly due to overuse of these agents by outpatient specialists and general practitioners. Complicating matters, resistance to cotrimoxazole may arise from overuse not only of cotrimoxazole but also amoxicillin. The previous study had shown that overuse of antimicrobial agents lead to development of resistant organisms. For instance, isolation of ciprofloxacin-resistant *Escherichia coli* strains collected from uncomplicated urinary tract infections were associated with usage of a fluoroquinolone in the preceding three months (Katsarolis *et al.*, 2008). At present, it appears that oral preparation of cefuroxime is the most suitable antimicrobial therapy for suspected Gram-negative infections in stable, non-admitted patients. Despite the increasing antibiotic resistance among Gram-negative microorganisms, this trend was not followed by their Gram-positive counterpart.

While typhoid fever is endemic in North-eastern Malaysia, the number of positive blood cultures reached a peak during an outbreak of typhoid in 2005. *Salmonella* Typhi, the causative agent of typhoid fever, was identified in 173 isolates in that year. That strain of *Salmonella* Typhi was sensitive to all antimicrobials tested, including ampicillin, cotrimoxazole and ciprofloxacin. Because patients may exhibit a wide range of non-specific symptoms, every emergency physician in this region should always take into account the possibility that a patient may be suffering from typhoid. The blood culture test for typhoid is most sensitive during the first week of illness, but some patients may not seek immediate medical attention. The range in the positivity rate among cultures from children was 50-70% (Choo *et al.*, 1994). The cultures normally took more than two days to provide a result, so an emergency physician should consider requesting a rapid serology diagnosis of typhoid and initiate anti-typhoid medication without waiting for the blood culture results. The emergency physician is also responsible for notifying public health authorities to prevent further spread of the pathogen.

Because this is a retrospective study, the selection of cases for blood culture was based on the judgement of the managing emergency physician. The reliability of blood culture results from the ED setting is questionable, especially when indication for blood culture was not properly established. Studies from Europe and North America have demonstrated that blood cultures obtained in an ED have little influence over clinical management (Sturmann *et al.*,1996; Kelly 1998; Leonard & Beattie, 2003). A quick diagnosis of bacteraemia for an ED patient is critical to reduce the chances of communicability (Bates *et al.*, 1991) and to guide his or her clinical management (Vorwerk *et al.*, 2009). In our ED setting, isolates containing coagulase-negative staphylococci, Gram-positive rods or mixed-species growth were considered contaminants and were responsible for 2.7-

14.3% of positive samples. The high contamination rate in 2002 (14.3%) was possibly due to manual processing of blood cultures. After switching the culture processing method to the automated BACTEC system, annual contamination rates dropped (2.7-6.5%). This range of contamination values for our ED setting was comparable to or better than those ranges achieved by previous studies: 3.1-7.4% (Gander *et al.*, 2009); 4.2-6.4% (Smart *et al.*, 1993); and 8.0-24.0% (Madeo *et al.*, 2005).

A detailed understanding of the pathogens common to a region, including their respective antimicrobial sensitivity patterns, is extremely important for the proper management of sepsis cases by ED personnel. Our results indicate there is a growing trend among Gram-negative species encountered in the ED of increased antimicrobial resistance, particularly to those agents regularly used in an outpatient setting. This study also suggests that rational use of antibiotics in outpatient and general practitioner settings is imperative to control the emergence of resistant strains. Further studies are required to evaluate the attitudes of physicians in outpatient settings with respect to prescription of antimicrobial agents.

REFERENCES

Bates, D.W., Goldman, L. & Lee, T.H. (1991). Contaminant blood cultures and resource utilization: The true consequences of false-positive results. *Journal of American Medical Association* **265**: 365-369.

Choo, K.E., Oppenheimer, S.J., Ismail, A.B. & Ong, K.H. (1994). Rapid serodiagnosis of typhoid fever by Dot Enzyme Immunoassay in an Endemic Area. *Clinical Infectious Disease* **19**: 172-176.

Dellinger, R.P., Levy, M.M.., Carlet, J.M., Bion, J., Parker, M.M., Jaeschke, R., Reinhart, K., Angus, D.C., Brun-Buisson, C., Beale, R., Calandra, T., Dhainaut, J.F., Gerlach, H., Harvey, M., Marini, J.J., Marshall, J., Ranieri, M., Ramsay, G., Sevransky, J., Thompson, B.T., Townsend, S., Vender, J.S., Zimmerman, J.L. & Vincent, J.L. (2008). Surviving Sepsis Campaign: International guidelines for management of severe sepsis and septic shock. Critical Care Medicine **36**: 296-327.

Gander, R.M., Byrd, L., DeCrescenzo, M., Hirany, S., Bowen, M. & Baughman, J. (2009). Impact of blood cultures drawn by phlebotomy on contamination rates and health care costs in a hospital emergency department. *Journal of Clinical Microbiology* **47**: 1021-1024.

Katsarolis, I., Poulakou, G., Athanasia, S., Kourea-Kremastinou, J., Lambri, N., Karaiskos, E., Panagopoulos, P., Kontopidou, F.V., Voutsinas, D., Koratzanis, G., Kanellopoulou, M., Adamis, G., Vagiakou, H., Perdikaki, P., Giamarellou, H. & Kanellakopoulou, K. (2008). Acute uncomplicated cystitis: from surveillance data to a rationale for empirical treatment. *Journal Antimicrobial Chemotherapy* **35**: 62-67.

Kelly, A.M. (1998). Clinical impact of blood cultures taken in the emergency department. *Journal of Accident & Emergency Medicine* **15:** 254-256.

Kumar, A., Ellis, P., Arabi, Y., Roberts, D., Light, B., Parrillo, J.E., Dodek, P., Wood, G., Kumar, A., Simon, D., Peters, C., Ahsan, M. & Chateau, D. (2009). Initiation of inappropriate antimicrobial therapy results in a fivefold reduction of survival in human septic shock. *Chest* **136**: 1237-1248.

Leonard, P. & Beattie, T.F. (2003). How do blood cultures sent from a paediatric accident and emergency department influence subsequent clinical management? *Emergency Medicine Journal* **20**: 347-348.

Madeo, M., Jackson, T. & Williams, C. (2005). Simple measures to reduce the rate of contamination of blood cultures in Accident and Emergency. *Emergency Medicine Journal* **22**: 810-811.

Martin, D.R. (2008). Preface. *Emergency Medicine Clinical North America Journal* **26**: 17-18.

Ramos, J.M., Masiá, M. & Elía, M. (2004). Epidemiological and clinical characteristics of occult bacteremia in an adult emergency department in Spain: influence of blood culture results on changes in initial diagnosis and empiric antibiotic treatment. *Euro Journal Clinical Microbiology Infectious Dieases* **23**: 881-887.

Schneider, J.I. (2004). Rapid infectious killers. *Emergency Medicine Clinical North America Journal* **22**: 1099-1115.

Smart, D., Baggoley, C., Head, J., Noble, D., Wetherall, B. & Gordon, D.L. (1993). Effect of needle changing and intravenous cannula collection on blood culture contamination rates. *Annals Emergency Medicine* **22**: 1164-1168.

Strehlow, M.C., Emond, S.D., Shapiro, N.I., Pelletier, A.J. & Camargo, J.C.A. (2006). National Study of Emergency Department Visits for Sepsis, 1992 to 2001. *Annals Emergency Medicine* **48**: 326-326.

Sturmann, K.M., Bopp, J., Molinari, D., Akhtar, S. & Murphy, J. (1996). Blood cultures in adult patients released from an urban emergency department: a 15-month experience. *Academy Emergency Medicine* **3**: 768-775.

Talan, D.A., Moran, G.J. & Abrahamian, F.M. (2008). Severe Sepsis and Septic Shock in the Emergency Department. *Infectious Disease Clinical North America* **22**: 1-31.

Vorwerk, C., Loryman, B. & Coats, T.J. (2009). Prediction of mortality in adult emergency department patients with sepsis. *Emergency Medicine Journal* **26**: 254-258.