

Trends in Bloodstream Infections at a Korean University Hospital between 2008 and 2013

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Background: Blood culture remains the definitive method for diagnosing bacteremia and fungemia. In this study, we investigated the incidence of bacterial and fungal infections along with the trends in antimicrobial susceptibility in blood cultures collected from 2008 to 2013.

Methods: We performed a retrospective analysis of blood cultures performed at Kyung Hee University Hospital, Seoul, South Korea, between 2008 and 2013 to determine the bacterial and fungal species isolated, and their antimicrobial susceptibilities. Additional analyses were performed comparing these results to that of a prior study examining blood cultures collected from 2003-2007.

Results: Of the 102,257 specimens collected, 8,452 (8.3%) were culture positive, with *Staphylococcus epidermidis* being the most common species isolated (17.3%), followed by *Escherichia coli* (16.9%), *Staphylococcus aureus* (8.1%), and *Klebsiella pneumoniae* (6.5%). Fungal species accounted for 3.7% of all iso-

lates. Methicillin resistance was seen in 54.3% of *S. aureus* isolates. The frequencies of extended-spectrum β -lactamase (ESBL)-producing *E. coli* and *K. pneumoniae* were 13.1% and 10.3%; imipenem resistance was seen in 19.5% of *Pseudomonas aeruginosa* isolates.

Conclusion: Although the number of blood specimens analyzed increased steadily over the course of this study, the rate of positive blood cultures declined. The most common microorganisms isolated were coagulase-negative staphylococci, *E. coli*, *S. aureus*, and *K. pneumoniae*, consistent with our prior analysis. This analysis of blood culture isolate frequencies and antibiotic susceptibilities can be used to inform antibiotic therapy decisions. (*Ann Clin Microbiol* 2015;18: 14-19)

Key Words: Antimicrobial susceptibility, Bacteremia, Bloodstream infections

INTRODUCTION

Bloodstream infections are a major cause of mortality in hospitalized patients, particularly among patients developing sepsis as a result of the infection. In the United States alone, more than 750,000 cases of sepsis occur every year, with a mortality rate close to 30% [1]. Early detection of the infectious agent, followed by appropriate antimicrobial therapy, represents the best way to control costs and improve patient outcomes. Despite recent advances in clinical diagnostics, blood culture remains the gold standard for the detection of bacteremia and fungemia. Analysis of the long-term trends in both infectious agents and

antimicrobial susceptibilities can be used to improve patient care, leading to reduced morbidity and mortality associated with these infections. Here, we performed a retrospective analysis of blood culture results collected over the course of 6 years. These results were then compared to that of a similar study examining blood cultures collected from 2003-2007 [2] to identify changes in infectious organisms and antimicrobial susceptibilities over time.

MATERIALS AND METHODS

A retrospective analysis of blood cultures performed at

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Kyunghee University Hospital, Seoul, South Korea between January 2008 to December 2013 was performed, examining the frequencies of bacterial isolates, and their respective antimicrobial susceptibilities. The data presented here include both true infections as well as instances of contamination but exclude overlapped isolates from identical patient.

1. Blood cultures

Aerobic blood cultures for adult and pediatric patients were analyzed with the Bactec 9120 and 9240 systems using BACTEC plus aerobic/F Medium and BACTEC peds plus/F culture vials (Becton Dickinson, Sparks, MD, USA). Culture-B medium (Hanil Komed, Sungnam, Korea) was used for anaerobic blood cultures collected from adults; anaerobic cultures were not conducted for pediatric patients. For adults, 5-10 mL of blood were inoculated into aerobic and anaerobic vials, respectively, and cultured at 35°C for 5 days; for pediatric patients, this volume was reduced to 1-2 mL of blood. Anaerobic cultures were manually checked daily for 5 days. Whenever a positive signal was detected, subculturing was performed to identify the infecting microorganism and to perform antimicrobial susceptibility testing.

2. Species identification and antimicrobial susceptibility testing

Bacterial species identification and antimicrobial susceptibility testing were performed using Microscan LabPro (Siemens Healthcare Diagnostics, West Sacramento, CA, USA); API 20NE and API 20E systems (bioMérieux, Durham, NC, USA) were also used, as necessary. With Microscan LabPro, identification of clinical isolates based on over 85 percent match. The Clinical and Laboratory Standard Institute (CLSI) guidelines were used to interpret antimicrobial susceptibilities [3]. Fungi were identified by culturing for at least 4 weeks on Sabouraud-

dextrose agar. CHROMagar Candida (CHROMagar, Paris, France) and germ-tube test were used to identify *Candida* species. Antifungal susceptibility was not tested.

3. Statistics analysis

The correlation analysis between year and frequencies of isolates (or antimicrobial susceptibilities) was performed using MedCalc for Windows, version 13.2.2.0 (MedCalc Software, Ostend, Belgium). A *P* value less than 0.05 was considered to be statistically significant.

RESULTS

Of the 102,257 blood specimens collected, 8,452 (8.3%) were culture positive for growth of microorganisms. Excluding overlapped isolates from identical patient, a total of 4,768 isolates were analyzed. Of these, 2,683 (56.3%) were aerobic gram-positive cocci and 1,601 (33.6%) were aerobic gram-negative bacilli; fungal species accounted for an additional 178 (3.8%) (Table 1).

1. Gram-positive cocci

The most common species of Gram-positive bacterium was *Staphylococcus epidermidis*, which accounted for 824 isolates (17.3%). Coagulase-negative staphylococci (CoNS) were identified in 1,768 isolates (37.1%). Excluding CoNS, *Staphylococcus aureus* was the most common species with 385 isolates (8.1%). Other common Gram-positive species identified in this study included *Enterococcus faecalis*, *Enterococcus faecium*, and *Streptococcus pneumoniae*, which accounted for 95 (2.0%), 91 (1.9%), and 44 (0.9%) isolates, respectively (Table 2).

Table 1. Classes of bacteria and fungi isolated by year

Organisms	Number of isolates (%) by year						2008-2013 Total	2003-2007 Total	[2]
	2008	2009	2010	2011	2012	2013			
Aerobic and facultative									
Gram-positive cocci	544	495	465	413	418	348	2,683 (56.3)	1,976 (59.8)	
Gram-negative cocci	3	0	0	5	1	0	9 (0.2)	5 (0.2)	
Gram-positive bacilli	35	35	52	42	50	63	277 (5.8)	90 (2.7)	
Gram-negative bacilli	261	252	268	254	281	285	1,601 (33.6)	1,095 (33.2)	
Anaerobic bacteria	0	4	3	3	3	6	19 (0.4)	1 (0.03)	
Fungi	29	28	30	33	28	30	178 (3.7)	136 (4.1)	
Undefined	0	1	0	0	0	0	1 (0.02)	0 (0.0)	
Total	872	815	818	750	781	732	4,768 (100)	3,303 (100)	

Table 2. Significant bacterial isolates by year

Organisms	Number of isolates (%) by year						2008-2013 Total	2003-2007 Total [2]
	2008	2009	2010	2011	2012	2013		
Gram-positive cocci								
<i>Staphylococcus</i> , coagulase negative	372 (42.7)	330 (40.5)	309 (37.8)	263 (35.1)	261 (33.4)	233 (31.8)	1,768 (37.1)	1,159 (36.6)
<i>S. aureus</i>	71 (8.1)	69 (8.5)	71 (8.7)	59 (7.9)	64 (8.2)	51 (7.0)	385 (8.1)	430 (13.6)
<i>Enterococcus faecalis</i>	14 (1.6)	21 (2.6)	14 (1.7)	15 (2.0)	18 (2.3)	13 (1.8)	95 (2.0)	72 (2.3)
<i>Enterococcus faecium</i>	24 (2.8)	17 (2.1)	16 (2.0)	10 (1.3)	18 (2.3)	6 (0.8)	91 (1.9)	95 (3.0)
<i>Streptococcus pneumoniae</i>	11 (1.3)	12 (1.5)	7 (0.9)	6 (0.8)	4 (0.5)	4 (0.6)	44 (0.9)	38 (2.0)
<i>Streptococcus agalactiae</i>	6 (0.7)	2 (0.2)	7 (0.9)	12 (1.6)	6 (0.8)	3 (0.4)	36 (0.8)	NA
<i>Streptococcus pyogenes</i>	3 (0.3)	2 (0.2)	0 (0.0)	1 (0.1)	3 (0.4)	1 (0.1)	10 (0.2)	NA
Gram-negative bacilli								
<i>Escherichia coli</i>	121 (13.9)	124 (15.2)	124 (15.2)	138 (18.4)	156 (20.0)	144 (19.7)	807 (16.9)	516 (16.3)
<i>Klebsiella pneumoniae</i>	42 (4.8)	50 (6.1)	74 (9.1)	44 (5.9)	51 (6.5)	50 (6.8)	311 (6.5)	185 (5.8)
<i>Acinetobacter baumannii</i>	16 (1.8)	9 (1.1)	12 (1.5)	12 (1.6)	13 (1.7)	23 (3.1)	85 (1.8)	NA
<i>Pseudomonas aeruginosa</i>	21 (2.4)	14 (1.7)	14 (1.7)	17 (2.3)	15 (1.9)	6 (0.8)	87 (1.8)	62 (2.0)
<i>Enterobacter</i> spp.	7 (0.8)	8 (1.0)	9 (1.1)	9 (1.2)	8 (1.0)	11 (1.5)	52 (1.1)	58 (1.8)
<i>Serratia marcescens</i>	6 (0.7)	4 (0.5)	1 (0.1)	3 (0.4)	1 (0.1)	3 (0.4)	18 (0.4)	21 (0.7)
<i>Salmonella</i> Typhi	0 (0.0)	3 (0.4)	0 (0.0)	0 (0.0)	1 (0.1)	1 (0.1)	5 (0.1)	6 (0.2)
Anaerobic bacteria								
<i>Clostridium perfringens</i>	0 (0.0)	4 (0.5)	1 (0.1)	2 (0.3)	1 (0.1)	3 (0.4)	11 (0.2)	1 (0.0)
Total	714 (81.9)	669 (82.1)	659 (80.8)	591 (78.9)	620 (79.3)	552 (75.3)	3,805 (79.8)	2,643 (84.3)

Abbreviation: NA, not available.

Table 3. Fungal isolates trend

Organism	Number of isolates (%)	
	2008-2013	2003-2007 [2]
<i>Candida albicans</i>	74 (42.0)	55 (40.4)
<i>Candida tropicalis</i>	63 (35.0)	47 (34.6)
<i>Candida parapsilosis</i>	22 (12.0)	12 (8.8)
<i>Candida glabrata</i>	13 (7.3)	15 (11.0)
Other	6 (3.4)	7 (5.2)
Total	178 (100)	136 (100)

2. Gram-negative bacilli

The most common species of Gram-negative bacillus was *Escherichia coli*, which was identified in 807 isolates (16.9%). Other common Gram-negative pathogens included *Klebsiella pneumoniae* (311 isolates; 6.5%), *Pseudomonas aeruginosa* (87 isolates; 1.8%), and *Acinetobacter baumannii* (85 isolates; 1.8%) (Table 2).

3. Fungi and anaerobic bacteria

One hundred and seventy-eight fungal isolates were identified, the most common being *Candida albicans* (74 isolates) and *Candida tropicalis* (63 isolates) (Table 3). The most common

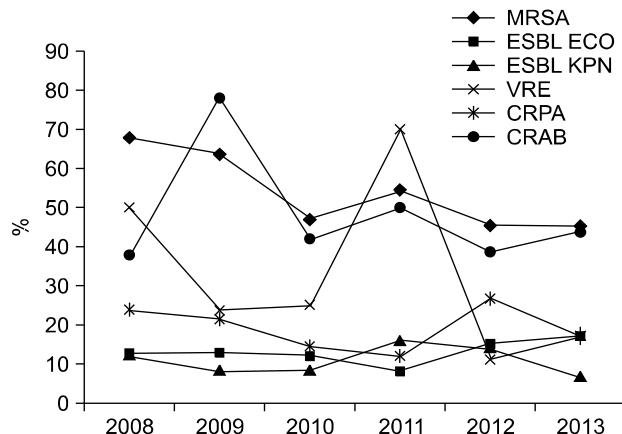


Fig. 1. Trends in antimicrobial resistance rates in *S. aureus*, *E. coli*, *K. pneumoniae*, *E. faecium*, *P. aeruginosa* and *A. baumannii*. Abbreviations: MRSA, Methicillin-resistant *S. aureus*; ESBL ECO, Extended-spectrum β-lactamase-producing *E. coli*; ESBL KPN, Extended-spectrum β-lactamase-producing *K. pneumoniae*; VRE, Vancomycin-resistant *E. faecium*; CRPA, Carbapenem-resistant *P. aeruginosa*; CRAB, Carbapenem-resistant *A. baumannii*.

anaerobic bacterial species was *Clostridium perfringens* (0.2%).

4. Antimicrobial susceptibility

Methicillin resistance was detected in 54.3% of *S. aureus* isolates; however, the rate exhibited a clear downward trend (Fig.

Table 4. Trend of vancomycin and ampicillin resistance in *E. faecalis* and *E. faecium*

	<i>E. faecalis</i>				<i>E. faecium</i>			
	Vancomycin		Ampicillin		Vancomycin		Ampicillin	
	R/Tested	%	R/Tested	%	R/Tested	%	R/Tested	%
2008	1/14	7.1	0/14	0	12/24	50.0	20/24	83.3
2009	0/21	0	0/21	0	4/17	23.5	16/17	94.1
2010	0/14	0	0/14	0	4/16	25.0	13/16	81.3
2011	0/15	0	0/15	0	7/10	70.0	10/10	100.0
2012	2/18	11.1	1/18	5.6	2/18	11.1	15/18	83.3
2013	0/13	0	0/13	0	1/6	16.7	3/6	50.0
Total	3/95	3.2	1/95	1.1	30/91	33.0	77/91	84.6

Abbreviation: R, resistant.

1). Resistance to vancomycin was detected in 33.0% and 3.2% of *E. faecium* and *E. faecalis* isolates, respectively (Table 4). Ampicillin resistance was common among *E. faecium* isolates, accounting for 84.6% of all isolates; resistance was also observed in *E. faecalis*, albeit at a considerably lower rate (1.1%) (Table 4). The frequencies of extended-spectrum β -lactamase (ESBL)-producing *E. coli* and *K. pneumoniae* were 13.1% and 10.3%, respectively. Imipenem and meropenem resistance was observed in 19.5% and 16.1% of *P. aeruginosa* isolates, respectively; higher rates of meropenem resistance were seen in *A. baumannii*, in 46.8% of the total number of isolates of this taxon (Fig. 1).

DISCUSSION

Despite a steady increase in the number of blood specimens collected annually, the rate of positive blood cultures decreased over the course of this study. Overall, blood cultures were positive in 8.3% of all blood specimens collected, a rate similar to that of our previous study (8.5%) of blood culture specimens collected between 2003 and 2007 [2]. The most common microorganisms identified were CoNS, *E. coli*, *S. aureus*, and *K. pneumoniae*, consistent with both our previous results as well as that of similar studies [2,4-7]. Antibiotic resistance rates varied considerably from year to year.

Among the most notable findings of this study was the steady change in the isolation rates of two common pathogens: CoNS and *E. coli*. CoNS isolation rates decreased by 10.9 percentage points over the course of 6 years; in contrast, the rate of *E. coli* isolation increased by 5.8 percentage points (Table 2). Between 2008 and 2010 the most common species isolated from blood

cultures was *S. epidermidis*; in the years since, *E. coli* has emerged as the most common isolate.

CoNS are generally regarded as contaminants, the majority of which are picked up from the patient's skin during the process of blood collection [8]. In this study, the reason for decrease of CoNS isolation is also considered to be due to reduced contaminants. However, CoNS are also frequent nosocomial pathogens causing severe infections in conjunction with intra-vascular devices such as central venous catheters [8,9]. Therefore, identification of these pathogens at the species level is recommended, followed in cases of repeated isolation by antibiotic susceptibility testing.

Other than CoNS, *S. aureus* was the most common species isolated from blood. The frequency of *S. aureus* isolation reported here represents a modest decrease (10.5%) compared to our previous study (Table 2). The rate of methicillin resistance also exhibited a downward trend in this cohort ($P<0.05$); more modest decreases in vancomycin resistance were observed among *E. faecium* isolates, while the rate of ampicillin resistance had increased relative to our earlier study [10].

E. coli isolation rates exhibited modest, but consistent, increased over the course of this study, while *K. pneumoniae* frequencies remained constant, with isolation rates similar to in our previous study. Increase of proportion and absolute value of *E. coli* isolation is thought to be due to increase of patients or severity of patients. The isolation frequencies of ESBL-producing *E. coli* and *K. pneumoniae* were 13.1% and 10.3%, respectively, and remained relatively stable from year to year. Due to increasing use of carbapenems for empiric therapy, carbapenemase-producing *Enterobacteriaceae* (CPE) strains have spread rapidly [11]. We identified two *E. coli* isolates (both from 2010) re-

sistant to meropenem (data not shown).

Both *P. aeruginosa* and *A. baumannii* were identified in 1.8% of culture-positive cases. However, the rate of meropenem-resistant *A. baumannii* was approximately threefold higher than that of meropenem-resistant *P. aeruginosa*. *A. baumannii* is an important opportunistic pathogen responsible for nosocomial infections, commonly in association with intravascular catheters and in immunocompromised patients. Rigorous infection control practices are necessary in cases of *A. baumannii* infection, as multidrug or pandrug-resistant *A. baumannii* strains are commonplace.

The frequencies of methicillin-resistant *S. aureus* and carbapenem-resistant *A. baumannii* represented noticeable decreases in 2010. These declines seem to be a consequence of enhanced infection control practices in 2010, but other species like *E. coli* and *E. faecium* exhibited various trends.

While the isolation frequencies of anaerobic bacteria exhibited no significant differences with our previous results, the rate was relatively low compared to that of similar studies [12,13]. The utility of manual detection of positive vials is considered unsuitable.

In addition to bacteria, 178 fungal isolates were identified. The most frequently isolated fungus was *C. albicans*, followed by *C. tropicalis*. However, the frequency of *C. tropicalis* isolation decreased rapidly over the course of this study, such that it was overtaken by *C. parapsilosis* and equal to that of *C. glabrata* by 2013. Due to absence of antifungal susceptibility data, the correlation between changing isolation trends of *Candida* spp. and known resistant pattern did not represent. As systemic fungal infections are associated with high mortality [14], rapid detection in conjunction with appropriate antifungal therapy is essential to ensure a positive outcome.

As with all research, this study was not without limitations. First, the data presented here include both true infections as well as instances of contamination. Second, identification of clinical isolates with Microscan LabPro based on over 85% match, so some identification of species level could be inaccurate. Third, we were forced to exclude a portion of the antimicrobial susceptibility testing results, due to the absence of interpretation criteria. More detailed laboratory and patient records are necessary to avoid these issues in any subsequent analyses. Fourth, the total number of antimicrobial susceptibilities testing is somewhat scanty, so the rate of resistance can be easily changeable.

In conclusion, we detected a consistent decrease in the frequency of CoNS isolation over the course of this study, com-

bined with a steady increase in the frequency of isolation of *E. coli*. Approximately 50% of *A. baumannii* isolates exhibited significant carbapenem resistance (imipenem or meropenem). Comprehensive and strict infection control practices will be necessary to reduce the rate of nosocomial infections going forward. Combined with our previous efforts, despite results of single hospital, this study represents a long-term analysis of blood stream infections that can be used to identify trends in microbial pathogenesis and drug resistance. These data can be used to improve patient care, as well as reduce the costs associated with bloodstream infections.

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=국문초록=

2008년부터 2013년까지 5년간 한 대학병원에서의 혈액 배양 세균 추세

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배경: 혈액 배양은 균혈증과 진균혈증의 진단에 결정적인 방법이다. 본 연구에서 저자들은 2008년부터 2013년까지 시행된 혈액 배양에서 세균과 진균의 감염률과 항균제 감수성의 추세를 분석하였다.

방법: 2008년부터 2013년까지 혈액에서 분리된 세균, 진균, 그리고 세균의 항균제 감수성 결과를 확인하기 위하여 6년간 본원에서 시행된 혈액 배양 결과를 후향적으로 분석하였다. 추가적으로 2003년부터 2007년까지의 혈액 배양 결과의 이전 연구와 비교 분석하였다.

결과: 총 102,257검체 중 8,452 (8.3%)가 양성이었으며, *Staphylococcus epidermidis*가 가장 흔히 분리되었고(17.3%), 이어 *Escherichia coli* (16.9%), *Staphylococcus aureus* (8.1%), 그리고 *Klebsiella pneumoniae* (6.5%) 순으로 분리되었다. 진균은 전체 분리균주의 3.7%였다. Methicillin 내성은 *S. aureus* 분리균주의 54.3%에서 관찰되었으며, Extended-spectrum β -lactamase (ESBL) 생성 *E. coli* and *K. pneumoniae*는 각각 13.1%, 10.3%였고, 19.5%의 *Pseudomonas aeruginosa* 균주에서 imipenem 내성이 관찰되었다.

결론: 혈액 검체의 수는 해마다 지속적으로 증가하였으나 양성률은 감소하였다. 가장 흔히 분리되는 미생물은 coagulase-negative staphylococci, *E. coli*, *S. aureus*, *K. pneumoniae*이었고, 본 연구의 이전 결과와도 일치하였다. 혈액 배양에서 균주 분리 빈도와 항균제 감수성 결과의 분석은 치료적 항균제 결정에 정보를 제공할 것이라 생각된다. [Ann Clin Microbiol 2015;18:14-19]