INTRODUCTION

Bacteremia is defined as the presence of microorganisms in the bloodstream causing clinical illness with the possibility of sepsis [1]. It is one of the major causes of mortality and morbidity worldwide and requires early appropriate antibiotic treatment [2]. Patients suspected of having bacteremia are often treated with empirical antibiotics before the causative microorganism is identified. To ensure that an appropriate antibiotic treatment is given, the distribution of the major pathogens and antibiotic susceptibility patterns should be known. The analysis of the trends in the types of bacteria isolated and their antibiotic susceptibility patterns can be used to improve patient care, leading to reduced morbidity and mortality associated with bloodstream infections or sepsis [3, 4]. Changes in the causative microorganisms of bacteremia and their antimicrobial susceptibility patterns in countries outside of Korea have been reported several times through either the SENTRY Antimicrobial Surveillance Program or the European...
Antimicrobial Resistance Surveillance System [3-5]. There have been few reports on bacterial pathogens and antibiotic susceptibility patterns in Korea prior to 2013 [6-8]. The purpose of this study was to identify the types of isolated bacteria and their antibiotic susceptibility patterns in blood cultures obtained at a university hospital over the 10-year period from 2007 to 2016.

MATERIALS AND METHODS

This study was conducted at a 700-bed university hospital located in an urban area. Data from blood cultures collected between January 2007 and December 2016 were retrospectively obtained from Kangbuk Samsung Hospital, Seoul, Korea. Relevant data included the date of culture, department, diagnosis, symptoms of infection, specimen serial number, isolated organisms, number of positive blood cultures with the same organism, and results of susceptibility testing. To prevent the duplication of antimicrobial susceptibilities, duplicate strains were excluded when the blood culture from the same person was rechecked within 3 days of hospitalization.

1. Blood cultures

Blood specimens were collected and inoculated into blood culture bottles using BD BACTEC Plus Aerobic/F, Lytic/10, Anaerobic/F, and Peds Plus/F (Becton Dickinson, Sparks, MD, USA) between January 2007 and December 2016, and the BACTEC FX system (BD Diagnostics, Heidelberg, Germany) was used to monitor bacterial growth. For adults, 5–10 mL of blood was inoculated into both aerobic and anaerobic vials; for pediatric patients, this volume was reduced to 1–2 mL of blood. Cultured blood bottles were incubated for 5 days in an automatic blood culture machine.

2. Identification of microorganisms and antimicrobial susceptibility testing

When a signal-positive blood culture bottle was detected using the BACTEC FX system, an aliquot was taken from the bottle for Gram staining and subculture onto appropriate solid media. When subculturing was carried out, we observed the pattern of growth for 7–10 days. Isolated colonies were identified, and antimicrobial susceptibility testing (AST) was performed using conventional methods with the VITEK2 system (bioMérieux, Marcy l’Etoile, France) and according to the standards of the Clinical and Laboratory Standards Institute (CLSI) [9, 10]. Identification and AST cards were tested periodically for quality control assessment.

3. Statistical analysis

Correlations between year and frequencies of isolates and/or antimicrobial susceptibilities were analyzed using IBM SPSS, version 24.0 (IBM Co., New York, NY, USA), and Excel 2010 (Microsoft Co., Redmond, WA, USA).

RESULTS

A total of 203,651 blood specimens were collected from 84,171 patients during the study period (from January 2007 through December 2016). Of these, 9,529 specimens (4.7%) were culture-positive for the growth of microorganisms, and the number of patients with an infectious disease (among patients with culture-positive specimens) was 8,196. Among these patients, there were 7,029 gram-positive bacterial isolates (83.7%), 917 gram-negative bacterial isolates (13.3%), and 250 fungal isolates (3.0%).

1. Frequency of isolated organisms from blood cultures by year

During the analysis period, the number of isolates per year increased steadily, and the distribution of isolated organisms varied with time (Fig. 1). Coagulase-negative staphylococci (CoNS), the most commonly isolated species during the entire period, increased from 23.7% in 2007 to 32.4% in 2016, and the isolation rate of Acinetobacter baumannii also increased from 2.2% in 2007 to 2.4% in 2016. On the other hand, Escherichia coli and Staphylococcus aureus showed decreasing trends, from 27.4% and 6.7% in 2007 to 21.8% and 6.6% in 2016, respectively.

2. Frequency of isolated organisms from blood cultures by age

The results were analyzed by age group, and S. aureus (5.7%) and E. coli (4.6%) were isolated from the under 10 years old group (Table 1). They were also isolated from the 10–19 years old group, but the isolation rate of S. aureus was higher than that in the other groups. The highest rate for E. coli, 25.2% was from the 70–79 years old group. E. faecium and E. faecalis showed a tendency to increase with increasing age.
3. Frequency of isolated organisms from blood cultures by departments of medicine

Fig. 2 shows the distribution of isolated organisms from blood cultures between January 2007 and December 2016 by departments of medicine (the nine departments of medicine in which blood cultures were most frequently performed and a group labeled “others” in which all other departments of medicine with less than 20 blood cultures in 1 year were combined). There were 15 medical departments in the others group, including cardiovascular surgery, neurology, orthopedic surgery, urology, and rehabilitation medicine. The department of medicine with the highest frequency over the 10-year period was the emergency room (ER), followed by gastroenterology (MG) and medicine of oncology (MO). The most common isolates in the ER were E. coli (36.0%), followed by S. aureus and Staphylococcus epidermidis (10.7% and 9.7%, respectively). In the other medical departments, slight

<table>
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</table>
differences were observed in the isolated organisms, but the isolates were usually almost identical.

4. Frequency of isolated organisms from blood cultures

1) Gram-positive cocci

CoNS was identified in 2,645 isolates (32.3%) (Table 2). The most common gram-positive bacterial species was *S. epidermidis*, which accounted for 1,380 isolates (16.8%). Excluding CoNS, *Staphylococcus aureus* was the most common species, with 658 isolates (8.0%). Other common gram-positive species identified in this study were *Enterococcus faecium*, *Enterococcus faecalis*, and *Streptococcus pneumoniae*, which accounted for 227 (2.8%), 223 (2.7%), and 69 (0.8%) isolates, respectively.

2) Gram-negative bacilli

The most common species of gram-negative bacillus was *E. coli*, which was identified in 1,867 isolates (22.8%) (Table 2). Other common gram-negative species were *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Acinetobacter baumannii*, which accounted for 664 (8.1%), 178 (2.2%), and 177 (2.2%) isolates, respectively.

3) Fungi and anaerobic bacteria

*Candida albicans* was the most commonly isolated fungal species (94 isolates, 1.1%), followed by *Candida parapsilosis* (51 isolates, 0.6%) (Table 2). The most common anaerobic bacterial species was *Propionibacterium acnes* (99 isolates, 1.1%).

5. Antimicrobial susceptibility

Methicillin (oxacillin) resistance was detected in 55.2% of *S. aureus* isolates during the study. The rate of resistance among isolates exhibited a clear downward trend, from 66.7% in 2007 to 37.6% in 2014 (Fig. 3). However, resistance increased again to 64.4% in the last 2 years of the study period. Among the isolates
collected over the study period, 42.4% of *S. aureus* isolates were resistant to ciprofloxacin. Furthermore, 4.3% and 2.1% were resistant to rifampin and cotrimoxazole, respectively. During the study period, no vancomycin-resistant *S. aureus* isolates were identified.

Among the enterococci, 25.6% of *E. faecalis* isolates were resistant to ampicillin, while 89.4% of *E. faecium* isolates were resistant (Fig. 4). With a total of seven isolated strains over the 10-year study period, the rate of vancomycin-resistant *E. faecalis* was very low (i.e., less than 1–2 isolates, or 3.3–11.1%, per year). With a total of 52 isolated strains, the rate of vancomycin-resistant *E. faecium* ranged from 22.2% to 31.4% during the last 3 years of the study period. The rates of third-generation cephalosporin and fluoroquinolone resistance in *E. coli* showed increasing trends over time; in 2016, the rates of resistance were 25.8% and 37.1%, respectively (Fig. 5). The rates of third-generation cephalosporin and fluoroquinolone resistance in *K. pneumoniae* were less than 10% in 2007. They showed a sharp increase in 2010 and 2011 and then gradually decreased until 2013. In 2016, the rates of resistance were 20.0% and 12.6%, respectively.

The imipenem resistance rate of *P. aeruginosa* increased from 6.25% in 2007 to 20.0% in 2016 (Fig. 6). The ciprofloxacin resistance rate of *P. aeruginosa* was 12.5% in 2007. Resistance varied over the 10-year study period but did not change markedly. In
2009, the imipenem resistance and ciprofloxacin resistance rates of *A. baumannii* were 12.5% and 25.0%, respectively. Imipenem resistance gradually increased from 50.0% in 2007 to 65.4% in 2016, and ciprofloxacin resistance gradually increased from 58.3% in 2007 to 76.9% in 2016.

**DISCUSSION**

To effectively treat patients with bacteremia, it is important to have information about the distribution of pathogens commonly isolated from the blood cultures of hospitalized patients and their antibiotic susceptibility [2]. It has already been reported that the distribution of common pathogens causing bacteremia and their antibiotic susceptibility profiles may differ among similarly-sized medical institutions located in the same country [11].

In this study, among the 8,196 isolates reviewed, CoNS was the most commonly isolated species, followed by *E. coli*, *S. aureus*, and *K. pneumoniae*. Over time, the relative proportion of CoNS isolates gradually increased, while the relative proportion of *S. aureus* isolates decreased. *E. coli* and *K. pneumoniae* maintained similar patterns of abundance during the analysis period. Other reports that analyzed bacteremia in Korea used different analysis periods; however, CoNS represented the most frequently isolated organisms, followed by *E. coli* and *S. aureus* [7, 8].

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**Fig. 5.** Ten-year trend of percent resistance to third-generation cephalosporins and fluoroquinolone for *E. coli* and *K. pneumoniae*. Abbreviations: ECO, *E. coli*; KPN, *K. pneumoniae*; 3rd cepha, third-generation cephalosporins; FQ, fluoroquinolone; %R, % resistance.

**Fig. 6.** Ten-year trend of percent resistance to imipenem and fluoroquinolone for *P. aeruginosa* and *A. baumannii*. Abbreviations: PAE, *P. aeruginosa*; ABA, *A. baumannii*; IPM, imipenem; FQ, fluoroquinolone; %R, % resistance.
CoNS species have often been regarded as culture contaminants. However, increasing numbers of immunosuppressed patients and patients undergoing intravascular catheter procedures have resulted in an increase in the number of invasive infections caused by CoNS [12, 13]. It is difficult to interpret the meaning of the identification of CoNS in a blood culture [1, 13-15]. CoNS was identified in 2,645 isolates (32.3%) from our blood cultures. We anticipated that there would be a certain level of contamination by CoNS, but further investigations will be needed to explain the annual increase in the relative proportion of CoNS isolates.

The most frequently isolated bacteria from blood differ depending on the patient’s residence area and age. In the past, the most common Salmonella spp. isolated in Korea was Salmonella typhi, but the isolation rate of S. typhi decreased in the late 1970s [16, 17]. In this study, the rate of Salmonella spp. decreased from 0.6% in 2007 to 0.3% in 2016. A possible reason for this decrease is the improvement in environmental sanitation procedures and the relative increase in other bacteria, and similar results were reported for other university hospitals in Korea, although the study times differed [18].

The major glucose-nonfermentative gram-negative bacilli isolates were P. aeruginosa (2.2%), A. baumannii (2.2%), and Stenotrophomonas maltophilia (0.3%). A. baumannii is an opportunistic pathogen that survives well in the hospital environment, and its frequency is increasing recently. Specifically, patients in intensive care units are known to be at high risk for infection. Over the study period, the isolation rate of A. baumannii remained relatively constant from 2.2% to 2.4%, whereas the isolation rate of P. aeruginosa decreased from 3.0% to 1.0%. This result differed from the reported increase in glucose-nonfermentative gram-negative bacilli other than P. aeruginosa [19]. This decrease in bacterial isolation possibly indicates that the hospital thoroughly performs infection control measures.

The isolation rate of S. maltophilia was 0.2% in 2007, and it increased to 0.3–0.6% in 2011–2015 and decreased to 0.1% in 2016. S. maltophilia is an important hospital infection pathogen and has been implicated in a variety of opportunistic infections and sepsis in humans [20]. Particularly, in patients with impaired immune function, infection is often caused by longer lengths of stay and frequent use of intravascularly inserted artificial devices.

Fungal species were isolated from 3.0% of all positive blood cultures, and Candida spp. accounted for 95% of the fungi. The isolation rate of Candida albicans was 1.0% in 2007, but it gradually increased to 1.1% in 2016. In the United States, Candida spp. are the fourth most common cause of bacteremia in hospitals [21]. Candidiasis is an infectious disease with a high prevalence and mortality rate, which have increased significantly over the past 20 years [22, 23]. Moreover, fungemia has been reported in other studies (both in Korea and in other countries) and is dependent on several factors, such as the frequent use of antibiotics, invasive procedures, chemotherapy, and organ transplantation [7].

The isolation rate of S. aureus also increased over the study period. S. aureus was the second most frequently isolated gram-positive organism, and more than 60% of strains isolated in 2016 were identified as MRSA. Other studies have reported an increase in the rate of methicillin resistance, similar to the pattern observed in our results [24]. According to data from the Korean Nationwide Surveillance of Antimicrobial Resistance (KONSAR), MRSA remained at a constant level of 60–70% between 1997 and 2011 [25, 26]. Similarly, data from the Korean Antimicrobial Resistance Monitoring System (KARMS) revealed that a 60–75% MRSA level was reported from general hospitals between 2008 and 2015 [27].

When analyzed by age, the positive rate of blood culture was higher in patients over 50 years of age, possibly because of increases in the elderly population with chronic diseases and the number of immunocompromised patients. The most common bacteria isolated were E. coli, K. pneumoniae, and S. aureus. E. coli is an opportunistic pathogen that is found among the normal bacterial flora in the intestine, and it causes accidental infections in humans with impaired immune function. The age-based analysis revealed that the isolation rate increased as the age increased, reflecting the fact that as the elderly population vulnerable to infection increases, the numbers of patients with chronic illnesses and who are immunocompromised are increasing compared to levels in the past [28].

The department of medicine-based analysis revealed that the most common isolate was E. coli (66.1%) in the ER, and the isolation rate of Salmonella spp. was higher than that in other medical departments (56.4%). Specifically, S. pneumoniae had the highest isolation rate in the ER (65%), suggesting that the incidence of community acquired pneumonia is increasing. During the early empirical antimicrobial treatment of community acquired pneumonia, the selection of inappropriate antimicrobial agents may
lead to failure of the initial treatment and worsening of disease progression, while the abuse of too broad a range of antimicrobial agents may cause the emergence and spread of antimicrobial resistant bacteria in the long term [29, 30].

As the widespread use of antibiotics has progressed, resistant bacteria have increased rapidly, making it difficult to select appropriate antibiotics. In particular, data on the status of antimicrobial resistance are very important when selecting appropriate therapeutic agents at this time of increased antimicrobial resistance.

The resistance rate of *E. faecium* to ampicillin was consistently high during the 10-year study period, remaining between 75% and 100%. The resistance rate of *E. faecium* to vancomycin increased by more than 10%. This change is likely associated with an increase in vancomycin use to treat cases of MRSA and infections caused by the Enterococci. Ampicillin and vancomycin resistance in *E. faecalis* has been reported to be 0-2% in other studies in Korea [27, 31], similar to the results of this study.

*E. coli* was isolated from 22.8% of bacteremia cases, and the rates of third-generation cephalosporin- and fluoroquinolone-resistant *E. coli* showed increasing trends over time. Third-generation cephalosporin and fluoroquinolone resistance in *K. pneumoniae* increased between 2007 and 2016, although resistance to these drugs was most prominent during 2010 and 2011. It is difficult to determine whether this was caused by a cluster of linked infections, by increased antibiotic usage, or by another unknown factor. Further studies are needed to elucidate these causes.

The rates of imipenem resistance in *P. aeruginosa* and imipenem and fluoroquinolone resistance in *A. baumannii* have shown increasing trends since 2007. In fact, the results reported in Korean journals and surveys indicate that there have been increasing trends since the early 2000s [7, 8, 27]. The rate of ciprofloxacin resistance in *P. aeruginosa* was 5.3-16.7% over our 10-year study period.

As with all research, this study had some limitations. First, all potential bacterial contaminants (such as CoNS, *Bacillus* spp., *Corynebacterium* spp., *Enterococcus* spp., *Micrococcus* spp., and *Propionibacterium* spp.) were included in the analysis without any clinical interpretation. Even with clinical information, it would have been difficult to distinguish between a true infection and contamination. Previous studies have documented the difficulties in distinguishing contaminants from true pathogens in clinical settings [32]. Second, we did not characterize the demographic data or disease severity for the patients included in the study. In addition, this study did not consider changes in clinical antibiotic usage when comparing trends of antimicrobial susceptibility. Third, we recently started monitoring the amount of blood used for culturing with the BD system. Although we previously recommended using 5-10 mL for adults and 1-2 mL for pediatric patients, we could not monitor the amounts that were used.

In conclusion, we analyzed different types of bacterial isolates, relative isolation frequencies, and antibiotic susceptibility patterns in blood cultures obtained over a 10-year period in order to identify changes and trends. Among the isolates, CoNS was the most common, followed by *E. coli* and *S. epidermidis*. Isolation rates for MRSA, VRE, and CRPA showed different patterns when analyzed by year. Even though all our results are from a single hospital, our findings represent a long-term analysis of bloodstream infections that can be used to identify trends in the microorganisms isolated and their drug resistance. These results provide meaningful information on the changes in the frequency of isolated organisms and antimicrobial susceptibility for the study period of 2007-2016, a period for which there are no KONSAR data. This analysis of blood culture isolate frequencies, along with the results of antibiotic susceptibility testing, will help determine the appropriate antibiotic therapy for current and future patients.

요 약

배경: 혈액 배양은 균혈증을 일으키는 병원균을 확인하고 항생제 치료를 위한 균주를 깨는 중요한 검사이다. 본 연구에서는 혈액 배양을 통한 균 동정과 항생제 감수성 검사 결과의 연간 변화 양상을 살펴보았다.


결과: 10년의 연구 기간 동안 총 203,651건의 혈액 배양 검사가 시행되었다. 이 중에 그람 양성 구균은 2.15%, 그람 음성 간균은 0.55%, 진균은 0.12%에서 검출되었다. *Escherichia coli*가 22.8%로 가장 많았고, *Staphylococcus epidermidis* (16.8%) , *Klebsiella pneumoniae* (8.1%), *Staphylococcus aureus* (8.0%) 순이었다. 진균은 양성 혈액 배양 동정 중의 3.0%였으며, *Candida albicans* (1.1%)가 가장 흔한 품종으로 *Candida parapsilosis* (0.6%)가 그 뒤를 이었다. *S.aureus*의 55.2%에서 Meticillin내성성이 나타났다. Vancomycin내성 *Enterococcus* (VRE)와 carbapenem내성 *Pseu-
*domonas aeruginosa* (CRPA)의 비도는 각각 13.1%와 10.9%였다. MRSA, VRE 및 CRPA의 분리율은 매년 다른 양상을 보였다.

결론: 분리균주 중 *E.coli*가 가장 흔히 분리되었으며 *S. epidermidis, K. pneumoniae* 등의 균주도 뒤를 이었다. 오랜 기간 동안의 혈류 감염을 분석한 본 연구를 통하여 급성환주로 입으키는 병원 균종과 약제 감수성 결과와 양상을 파악해 볼 수 있었다.

**AUTHORS’ DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST**

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