

# The prevalence of resistant Gram-negative bacteraemia among hospitalized patients in Tucson, Arizona over a 12-month period; A retrospective single center study

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## Abstract

**Introduction:** The objectives of this retrospective review were to: (a) determine the prevalence of resistant Gram-negative bacteraemia among hospitalized patients; (b) evaluate antibiotic use; (c) determine the time taken for Gram staining to final species identification.

**Methods:** For this retrospective study, information was extracted from patients' electronic medical records. Eligible patients had been admitted to a 300-bed tertiary care hospital in Tucson, Arizona from October 2015 to October 2016, were over 18 years of age and had a positive blood culture for Gram-negative bacteraemia.

**Results:** In total, 84 patients with Gram-negative bacteraemia were identified; urinary tract infection was the most common source of infection (71%). ESBL-producing microorganisms were isolated from five (6%) patients and no MDR pathogens were identified. The median time to Gram stain was 20.5 hours and the median time to final identification was 54.5 hours. Delayed de-escalation of broad-spectrum antibiotics (i.e., >24 hours after final culture) occurred in 25% patients with a median length of hospital stay of 118 hours (range: 56–552 hours)

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compared with a median length of hospital stay of 89 hours (range: 5–334 hours) in the early de-escalation group.

**Conclusion:** The prevalence of bacteraemia due to resistant Gram-negative microorganisms is low (6%) in this institution. However, there may be room for improvement in the antimicrobial stewardship program with regard to rapid diagnostic testing.

### Keywords

Bacteraemia, Gram-negative, bloodstream infections, extended-spectrum beta-lactamases, multidrug resistance

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### Introduction

Despite the availability of effective antibiotic therapies, bloodstream infections are an important cause of morbidity and mortality.<sup>1</sup> Importantly, bacteraemia due to Gram-negative bacilli is associated with a poor outcome because of the emergence of antibiotic resistance to these pathogens.<sup>2</sup> Moreover, antimicrobial resistance is considered a growing problem worldwide.<sup>3</sup> For example, in Japan, the increased use of anti-pseudomonal antibiotics has been linked to an elevated risk of multidrug-resistant organisms (MDROs).<sup>4</sup> Therefore, in many countries, health care institutions have set up antimicrobial stewardship programs (ASP) in an attempt to optimize antimicrobial use among hospitalized patients to ensure cost-effective outcomes, avoid inappropriate use of antibiotics and decrease or avoid complications.<sup>5</sup>

Resistance of Gram-negative bacilli to antibiotics can be caused by one or more of the following mechanisms: production of beta-lactamases, porin loss, target modification and drug efflux.<sup>6</sup> The production of extended-spectrum beta-lactamases (ESBL) by Gram-negative bacteria, is of growing concern because they are resistant to all  $\beta$ -lactam antibiotics apart from carbapenems

and cephamycins.<sup>7</sup> ESBL-producing Enterobacteriaceae pose a significant threat not only to hospitalized patients but also to those in community settings.<sup>8</sup> Several risk factors have been reported to increase the risk of ESBL acquisition and include, prior exposure to beta-lactam antibiotics, length of hospital stay, residence in a long-term care facility and  $\geq 65$  years of age.<sup>9</sup> However, the early initiation of appropriate and effective antibiotic use within 72 hours has been linked to a decreased mortality risk due to multidrug resistant pseudomonal species.<sup>10,11</sup>

Standard techniques for microbiological identification are based on phenotypic methods, which require 48-72 hours for identification and antimicrobial susceptibility compared with rapid diagnostics tests which only need a few hours following bacterial growth.<sup>12</sup> For these tests, positive blood cultures are tested using molecular methods to determine causative pathogens or both causative pathogens and resistance markers.<sup>13</sup> The use of rapid diagnostic methods coupled with appropriate therapeutic interventions should improve the overall clinical outcome and mortality.<sup>13</sup>

The objectives of this retrospective review were to (a) determine the prevalence of ESBL-producing Enterobacteriaceae bacteraemia and MDRO bacteraemia

among hospitalized patients; (b) evaluate their antibiotic use (i.e., broad spectrum antibiotics, dose escalation/ and (c) determine the time taken for Gram staining to final species identification.

## Methods

This retrospective study included all patients over 18 years of age with positive blood culture for Gram-negative bacteraemia admitted to a 300-bed tertiary care hospital in Tucson, Arizona from October 2015 to October 2016. There were no exclusions.

Patient data were collected from the hospital's electronic medical records system (Cerner®). Demographic characteristics, source of infection, length of hospitalization, isolated microorganisms, time of sample collection, time of Gram stain, time of final culture sensitivities and susceptibilities, results of final sensitivities and susceptibilities, first empiric antibiotics used, timing of antibiotic administration, time on broad-spectrum antibiotics and time to de-escalate narrow-spectrum antibiotics were extracted from the database.

The following definitions were used:

- time to Gram stain = time between sample collection and first Gram stain results
- time to final culture sensitivities and susceptibilities = time between sample collection and final culture sensitivities and susceptibilities
- time to de-escalate = time to de-escalate the antibiotics to narrower spectrum after final culture sensitivities and susceptibilities.
- delayed de-escalation = >24 hours after final culture sensitivities and susceptibilities
- broad spectrum antibiotics = antibiotics with pseudomonal coverage or two injectable antibiotics with different spectra of activity
- first antibiotics use = any antibiotic used after sample collection but before final culture sensitivities and susceptibilities

Patients with ESBL-producing Enterobacteriaceae bacteraemia and MDRO bacteraemia had their characteristics (i.e., initial and direct antibiotic use and disposition) identified. MDRO was defined as non-susceptibility to at least one agent in three or more antimicrobial categories.<sup>14</sup> Patients who had antibiotics de-escalated in less than 24 hours were compared with those who had delayed de-escalation (i.e., >24 hours) in terms of resistant pathogens and length of hospital stay.

This was a quality improvement project as part of an antimicrobial stewardship rotation and was exempt from Institutional Review Board (IRB) approval.

## Statistical analyses

Data were analysed using IBM SPSS software (version 25.0 for Windows®; IBM Corp, Armonk, NY, USA). Normally distributed continuous variables were reported as means with standard deviation (SD). Non-normally distributed continuous variables were reported as medians with interquartile range (IQR) and Mann-Whitney *U* test was used to compare data where appropriate. A *P*-value <0.05 was considered to indicate statistical significance.

## Results

Eighty-four patients with Gram-negative bacteraemia were identified. Their demographics and baseline characteristics are shown in Table 1. Mean (SD) age of the cohort was 72 (15.9) years and most patients were older than 60 years (80%) and female (69%) In total, 17% patients had recently been hospitalised and 12% were in long-term care facilities. Approximately a quarter of the cohort had recently received antibiotics most of which were broad spectrum. The urinary tract (71.4%), followed by gastrointestinal tract (17.8%) were the most common sources of infection (Table 2).

**Table 1.** Demographics and baseline characteristics.

Baseline characteristics	Total (n = 84)
Age, years	72 ± 15.9
Age >60 years	67 (79.8)
Sex, male	26 (31.0)
Sex, female	58 (69.0)
Recent hospitalization	14 (16.7)
Long term care facility (LTCF)	12 (12.3)
Any recent antibiotics use	25 (29.8)
Recent exposure to broad spectrum antibiotics* (e.g., vancomycin, cephalosporins, piperacillin/tazobactam)	24 (28.6)

Data are presented as mean ± SD or n (%).

\*Antibiotics with pseudomonal coverage or two injectable antibiotics with different spectrums of activity.

**Table 2.** Sources of infection.

Sources of infection	Total (n = 84)
Urinary tract	60 (71.4)
Gastrointestinal	15 (17.8)
Pneumonia	2 (2.4)
Vascular catheter related infection	1 (1.1)
Cellulitis	2 (2.3)
Unknown/unclear	4 (4.7)

Data are presented as n (%).

In total, 83 microorganisms were isolated from the 84 patients. Multidrug resistant carbapenemase-producing *Providencia stuartii* was initially thought to be isolated from one patient but this was found to be negative after using a modified Hodge Test to perform phenotypic testing for carbapenemase activity. *Escherichia coli* was the most commonly isolated microorganism (56/83, 67.4%), followed by *Klebsiella pneumoniae* (13/83, 15.6%), *Enterobacter* species (5/83, 6%) and *Pseudomonas aeruginosa* (2/83, 2.4%) (Figure 1). No MDROs were identified but five (6%) ESBL-producing

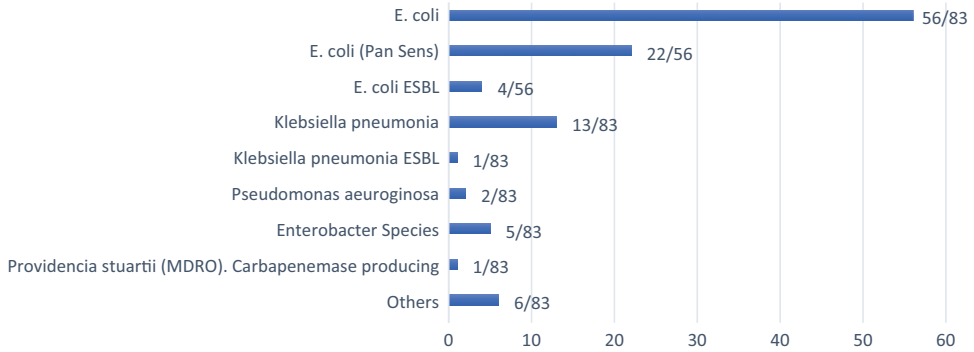
pathogens were identified (four *E. coli* and one *K. pneumoniae*).

All ESBL-infected patients were older than 60 years. Some details of these patients are shown in Table 3. Of the five patients, one had a recent hospitalization, three had a history of broad-spectrum antibiotic exposure and one was a long-term care facility resident. Four ESBL-infected patients had experienced delayed first effective antibiotic initiation; for three the source of infection was the urinary tract and for one patient it was unknown. Two were discharged home, one was discharged to a skilled nurse facility and one died.

Across the entire group, the median time to Gram stain was 20.5 hours (interquartile range 12.31 hours) and the median time to final culture sensitivities and susceptibilities was 54.5 hours (interquartile range 19 hours) (Figure 2). Therefore, the median time from gram stain to final culture was 29.4 hours (interquartile range 27.4 hours)

According to our pre-defined definition, antibiotics with antipseudomonal activity were used in 48/84 (57.1%) patients. This included one or more of the following: piperacillin-tazobactam, levofloxacin, meropenem, and aztreonam. Of the two patients who had *P. aeruginosa* bacteraemia, one received meropenem and vancomycin and was then de-escalated to meropenem following final blood culture sensitivities. The other patient received piperacillin-tazobactam upon admission, which continued throughout the hospital stay. Both patients had acquired their infections in the community; for one patient the source of infection was cellulitis and for the other patient it was a urinary tract infection. In our opinion, vancomycin had been used inappropriately in 10/84 (11.9%) patients with urinary source bacteraemia.

Delayed de-escalation of broad-spectrum antibiotics (i.e., >24 hours after final culture) occurred in 12/48 (25%) patients who had a median length of

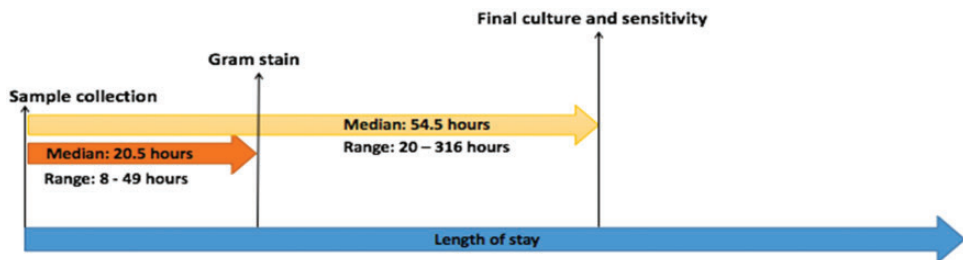


**Figure 1.** Isolated microorganisms (n = 83).

E. coli, *Escherichia coli*; Pan Sens, Pansensitive; ESBL, Extended-spectrum beta lactamases; Pan Sens: Pansensitive; MDRO: Multidrug-resistant organism; Others, *Citrobacter Freundii*, *Achromobacter xylosoxidans*, *Morganella morganii*, *Proteus mirabilis*, unspciated Gram-negative rod (2).

**Table 3.** Extended-spectrum beta-lactamases (ESBL)-infected patients (n = 5) and their characteristics.

Patient	Initial antibiotic	Directed antibiotic	Disposition
#1	Ceftriaxone	Meropenem	Home
#2	Levofloxacin + Piperacillin-tazobactam	Piperacillin-tazobactam	Deceased
#3	Ceftriaxone	Piperacillin-tazobactam	Deceased
#4	Levofloxacin	Meropenem	Home
#5	Levofloxacin + Piperacillin-tazobactam + Vancomycin	Piperacillin-tazobactam	Home



**Figure 2.** Time to Gram stain and final sensitivities.

hospital stay of 118 hours (range: 56–552 hours). Resistant pathogens were not identified in any of the delayed de-escalated group. By contrast, 33/48 (75%) of patients had their broad-spectrum antibiotics de-escalated appropriately (i.e., within 24 hours of final culture). The median length of hospital stay for these patients was 89 hours range: 5–334 hours. The

difference in hospital length of stay between delayed and early de-escalation groups was statistically significant ( $P$  0.02).

### Discussion

The prevalence of ESBL-producing Enterobacteriaceae differs among patients and geographic regions.<sup>15</sup> Moreover, rates

of ESBL-producing bacilli in *K pneumoniae* isolates tend to be higher than those in *E coli*.<sup>16</sup> For example, data from a global surveillance database showed the rate of ESBL production was highest among the *K. pneumoniae* isolates collected in Latin America, followed by Asia/Pacific Rim, Europe, and North America (44.0%, 22.4%, 13.3%, and 7.5%, respectively).<sup>15</sup> By contrast, data from a global study of the *in vitro* susceptibilities of *E. coli* strains isolated from individuals with urinary tract infections at 88 international hospitals, found that overall, 17.9% of isolates were ESBL producers with multidrug resistance. The highest rates were seen in Asia/ Pacific (27.7%), while the lowest rates were seen in North America (7.4%).<sup>17</sup> In addition, a retrospective study of data collected from five hospitals in the Netherlands during the period 2008 to 2010, found that of the 232 consecutive patients with ESBL bacteraemia due to *E coli*, and *K. pneumoniae* the ESBL producing prevalence among blood culture isolates of these bacteria were 6.6% and 8.7%, respectively.<sup>18</sup> Therefore, our findings of 6% bacteraemia caused by resistant Gram-negative microorganisms in our institution is reassuring and encourages us to continue with the adherence to national and local guidelines. In our study, ESBL-producing pathogens were identified in four *E. coli* and one *K. pneumoniae* isolates. These results are not surprising since urinary tract infections were common in this cohort and *E. coli* is the most important uropathogen.<sup>17</sup> In addition, while the U.S. Centers for Disease Control and Prevention (CDC) rated the level of carbapenem-resistant Enterobacteriaceae (CRE) as urgent<sup>1</sup> and data from a nationwide US Surveillance Network identified that CRE accounted for 4% of bloodstream infections,<sup>19</sup> no CRE was identified at our institution.

Timely administration of appropriate antibiotics is essential for decreasing

hospital mortality in patients with blood infections. For example, one study found that with each hour delay of effective antimicrobial therapy, there was a 7.6% decrease in survival in patients with sepsis.<sup>20</sup> Moreover, delaying appropriate antibiotic use can increase healthcare costs, length of stay and antimicrobial resistance.<sup>21,22</sup> In this present study, we identified only 4/84 patients with delayed initiation of effective antibiotics, and all had ESBL-producing pathogens. All of these patients had ESBL bacteraemia and only one patient died.

For traditional methods of microbial identification, the length of time taken for the detection of the pathogen is an important consideration. Most laboratories take 48–72 hours to complete the antimicrobial susceptibility testing.<sup>12,23</sup> This study showed that in our institution the median time from sample collection to final culture sensitivities and susceptibilities was 54.5 hours and from gram stain to final culture the median time was 29.4 hours. In addition, 75% of patients had their broad-spectrum antibiotics de-escalated appropriately (i.e., within 24 hours of final culture) and had a significantly shorter hospital stay than patients who had late de-escalation of antimicrobials. Therefore, similar to the findings of other studies,<sup>24</sup> our ASP is efficient and has led to decreased length of hospital stay. However, the introduction of rapid diagnostic testing at our institution may result in faster de-escalation times and avoid any unnecessary use of broad-spectrum antibiotics.<sup>13</sup> Nevertheless, data from a prospective, randomized, controlled trial over two years involving 1489 patients, found that although rapid bacterial identification and susceptibility testing led to earlier changes and a significant reduction in antibiotic use, it did not reduce mortality.<sup>23</sup> Similar findings were reported in another study that compared an ASP coupled with polymerase chain reaction (PCR)-based blood culture identification with ASP

and conventional organism identification or conventional organism identification (control) alone.<sup>25</sup> Although microorganism identification was significantly shorter for the PCR-blood culture identification group, there was no difference in mortality. However, significantly shorter times to de-escalate were observed in the PCR-blood culture identification group compared with the other groups. While results from a retrospective study involving 232 patients with ESBL bloodstream infections found that inappropriate antimicrobial agent within the first 24 hours was not associated with increased risk of mortality,<sup>18</sup> early identification of resistant microorganisms is imperative and more studies are needed, especially in patients with deep-seated infections.<sup>26</sup>

Our current study has some limitations. For instance, it was a retrospective single-centre study using a small sample size and so the findings may not be robust compared with data from other facilities that have higher rates of MDROs. Additionally, we did not assess the severity of disease nor did we include a control group. We were also unable to assess potential complications from the use of broad spectrum or multiple antibiotics. We defined delayed de-escalation as >24 hours after final sensitivities and susceptibilities rather than the time from the gram stain results and we did not evaluate whether doses were optimized based on minimum inhibitory concentration (MIC) results for specific microorganisms and antibiotics. Finally, we did not evaluate whether the treating physician or the ASP pharmacist performed the de-escalation.

Selecting the most appropriate and cost effective rapid diagnostic test is challenging given the wide range of commercially available diagnostic tools, some of which require labour-intensive clinical laboratory work with high complexity.<sup>12</sup> In addition, practitioner awareness of the usefulness of rapid

diagnostic testing may be limited. For example, a study found that only 60% of practicing physicians adjusted their patients' therapy based on rapid-blood-culture-identification results, and therefore, de-escalation of antimicrobial therapy was not optimal.<sup>27</sup> Although we observed a low prevalence of resistant Gram-negative pathogens, the median time from Gram stain to final culture was 29.4 hours and so the introduction of rapid diagnostics tests may be of beneficial value in reducing any unnecessary use of broad-spectrum antibiotics. While we did not examine the cost of therapy, the use of rapid diagnostic testing may have an impact on the overall cost, as well as decrease resistance rates.<sup>25</sup> A prospective study is required to determine the utility and possible cost reduction involved in the implementation of rapid diagnostic testing coupled with ASP in a large patient sample.

### Declaration of conflicting interest

The authors declare that there are no conflicts of interest.


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### References

1. Centers for Disease Control and Prevention (CDC). Antibiotic resistance threats in the

- United States (2013) (available at: <https://www.cdc.gov/drugresistance/threat-report-2013>).
2. Boucher HW, Talbot GH, Benjamin DK Jr, et al. 10 x '20 Progress—development of new drugs active against gram-negative bacilli: an update from the Infectious Diseases Society of America. *Clin Infect Dis* 2013; 56: 1685–1694.
  3. World Health Organization. (2014) Antimicrobial resistance: global report on surveillance. WHO (available at: <http://www.who.int/iris/handle/10665/112642>).
  4. Ohmagari N, Hanna H, Graviss L, et al. Risk factors for infections with multidrug-resistant *Pseudomonas aeruginosa* in patients with cancer. *Cancer* 2005; 104: 205–212.
  5. MacDougall C and Polk RE. Antimicrobial stewardship programs in health care systems. *Clin Microbiol Rev* 2005; 18: 638–656.
  6. Suarez CJ, Lolans K, Villegas MV, et al. Mechanisms of resistance to beta-lactams in some common Gram-negative bacteria causing nosocomial infections. *Expert Rev Anti Infect Ther* 2005; 3: 915–922.
  7. Magiorakos AP, Srinivasan A, Carey RB et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. *Clin Microbiol Infect* 2012;18:268–281.
  8. Sankar S, Narayanan H, Kuppanan S, et al. Frequency of extended-spectrum beta-lactamase (ESBL)-producing Gram-negative bacilli in a 200-bed multi-specialty hospital in Vellore district, Tamil Nadu, India. *Infection* 2012; 40: 425–429.
  9. Shaikh S, Fatima J, Shakil S, et al. Risk factors for acquisition of extended spectrum beta lactamase producing *Escherichia coli* and *Klebsiella pneumoniae* in North-Indian hospitals. *Saudi J Biol Sci* 2015; 22: 37–41.
  10. Su TY, Ye JJ, Hsu PC, et al. Clinical characteristics and risk factors for mortality in cefepime-resistant *Pseudomonas aeruginosa* bacteremia. *J Microbiol Immunol Infect* 2015; 48: 175–182.
  11. Leibovici L, Shraga I, Drucker M, et al. The benefit of appropriate empirical antibiotic treatment in patients with bloodstream infection. *J Intern Med* 1998; 244: 379–386.
  12. Kothari A, Morgan M and Haake DA. Emerging technologies for rapid identification of bloodstream pathogens. *Clin Infect Dis* 2014; 59: 272–278.
  13. Riedel S and Carroll KC. Early identification and treatment of pathogens in sepsis: molecular diagnostics and antibiotic choice. *Clin Chest Med* 2016; 37: 191–207.
  14. Livermore DM. Fourteen years in resistance. *Int J Antimicrob Agents*. 2012; 39(4):283-94–1596.
  15. Dhillon RH, Clark J. ESBLs: A Clear and Present Danger? *Crit Care Res Pract* 2012;2012:625170.
  16. Hague R. What is the threat from extended spectrum  $\beta$ -lactamase-producing organisms in children? *Arch Dis Child* 2011 Apr;96(4):325–327.
  17. Hoban DJ, Nicolle LE, Hawser S, et al. Antimicrobial susceptibility of global inpatient urinary tract isolates of *Escherichia coli*: results from the Study for Monitoring Antimicrobial Resistance Trends (SMART) program: 2009-2010. *Diagn Microbiol Infect Dis* 2011;70:507–511.
  18. Frakkig FN, Rottier WC, Dorigo-Zetsma JW, et al. Appropriateness of empirical treatment and outcome in bacteremia caused by extended-spectrum- $\beta$ -lactamase-producing bacteria. *Antimicrob Agents Chemother* 2013;57:3092–3099.
  19. Zilberberg MD, Shorr AF. Prevalence of multidrug-resistant *Pseudomonas aeruginosa* and carbapenem-resistant Enterobacteriaceae among specimens from hospitalized patients with pneumonia and bloodstream infections in the United States from 2000 to 2009. *J Hosp Med* 2013;8:559–563.
  20. Kumar A, Roberts D, Wood KE, et al. Duration of hypotension before initiation of effective antimicrobial therapy is the critical determinant of survival in human septic shock. *Crit Care Med* 2006; 34: 1589–1596.
  21. Lehmann LE, Herpichboehm B, Kost GJ, et al. Cost and mortality prediction using polymerase chain reaction pathogen detection in sepsis: evidence from three observational trials. *Crit Care* 2010; 14: R186.
  22. Fraser A, Paul M, Almanasreh N, et al. Benefit of appropriate empirical antibiotic treatment: thirty-day mortality and duration



- of hospital stay. *Am J Med* 2006; 119: 970–976.
23. Kerremans JJ, Verboom P, Stijnen T, et al. Rapid identification and antimicrobial susceptibility testing reduce antibiotic use and accelerate pathogen-directed antibiotic use. *J Antimicrob Chemother* 2008; 61: 428–435.
  24. Perez KK, Olsen RJ, Musick WL, et al. Integrating rapid pathogen identification and antimicrobial stewardship significantly decreases hospital costs. *Arch Pathol Lab Med* 2013; 137: 1247–1254.
  25. MacVane SH and Nolte FS. Benefits of adding a rapid PCR-based blood culture identification panel to an established antimicrobial stewardship program. *J Clin Microbiol* 2016; 54: 2455–2463.
  26. Rodríguez-Baño J, Navarro MD, Retamar P, et al.  $\beta$ -Lactam/ $\beta$ -lactam inhibitor combinations for the treatment of bacteremia due to extended-spectrum  $\beta$ -lactamase-producing *Escherichia coli*: a post hoc analysis of prospective cohorts. *Clin Infect Dis* 2012;54:167–174.
  27. Donner LM, Campbell WS, Lyden E, et al. Assessment of rapid-blood-culture-identification result interpretation and antibiotic prescribing practices. *J Clin Microbiol* 2017; 55: 1496–1507.