Neither Blood Culture Positivity nor Time to Positivity is Associated with Mortality among Patients Presenting with Severe Manifestations of Sepsis: The FABLED Cohort Study

Katryn Paquette MD, David Sweet, Robert Stenstrom MD, Sarah N. Stabler Pharm D, Alexander Lawandi MD, Murtaza Akhter MD, Adam C. Davidson MD, Marko Gavric BSc, Rehman Jinah MD, Zahid Saeed MD, Koray Demir MD, Sassan Sangsari MD, Kelly Huang MD, Amirali Mahpour MD, Chris Shamatutu BSc, Chelsea Caya MSc, Jean-Marc Troquet MD, Greg Clark MD, Titus Wong MD, Cedric P. Yansouni MD, and Matthew P. Cheng MD, for the FABLED investigators

1 Division of Neonatology, Montreal Children’s Hospital, McGill University, 1001 Decarie Boulevard, Montreal, Quebec, H4A 3J1, Canada
2 Division of Critical Care Medicine, Vancouver General Hospital, University of British Columbia, 899 W 12th Ave, Vancouver, British Columbia, V5Z 1M9, Canada
3 Department of Emergency Medicine, St-Paul’s Hospital, University of British Columbia, 1081 Burrard Street, Vancouver, British Columbia, V6Z 1Y6, Canada
4 Department of Pharmacy Services, Surrey Memorial Hospital, 13750 96 Ave, Surrey, British Columbia, V3V 1Z2, Canada
5 Divisions of Infectious Diseases and Medical Microbiology, McGill University Health Center, McGill University, 1001 Decarie Boulevard, Montreal, Quebec, H4A 3J1, Canada
6 National Institutes of Health Clinical Center, Critical Care Department, 10 Center Drive Room 2C145, Bethesda, Maryland, 20892-1662, United States of America

© The Author(s) 2021. Published by Oxford University Press on behalf of Infectious Diseases Society of America.
This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivs licence (http://creativecommons.org/licenses/by-nc-nd/4.0/), which permits non-commercial reproduction and distribution of the work, in any medium, provided the original work is not altered or transformed in any way, and that the work is properly cited. For commercial re-use, please contact journals.permissions@oup.com
7 Department of Emergency Medicine, Maricopa Integrated Health Center, 2601 East Roosevelt Street, Phoenix, Arizona, 85008, United States of America
8 Department of Emergency Medicine, Lion’s Gate Hospital, 231 15th St E, North Vancouver, British Columbia, V7L 2L7, Canada
9 Department of Pulmonary and Critical Care Medicine, University of Arizona College of Medicine, 475 N 5th St, Phoenix, Arizona, 85004, United States of America
10 Department of Medicine, McGill University, 1001 Decarie Boulevard, Montréal, Québec, H4A 3J1, Canada
11 Department of Emergency Medicine, Vancouver General Hospital, University of British Columbia, 899 W 12th Ave, Vancouver, British Columbia, V5Z 1M9, Canada
12 Division of Respirology, University of Western Ontario, London Health Sciences Center, 1151 Richmond Street, London, Ontario, N6A 4V2, Canada
13 Faculty of Medicine, University of British Columbia, 317 – 2194 Health Sciences Mall, Vancouver, British Columbia, V6T 1Z3, Canada
14 Department of Medicine, McGill University, 1001 Decarie Boulevard, Montréal, Québec, H4A 3J1, Canada
15 Department of Emergency Medicine, McGill University Health Centre, 1001 Decarie Boulevard, Montréal, Québec, H4A 3J1, Canada
16 Department of Medical Microbiology, Vancouver Coastal Health, 899 W 12th Ave, Vancouver, British Columbia, V5Z 1M9, Canada
TAKE HOME MESSAGE: Neither blood culture positivity nor its time to positivity are significant predictors of mortality among patients with severe manifestations of sepsis. Our findings support the ongoing use and research into bundles of care aimed at treating both the infectious and inflammatory aspects of sepsis.
ABSTRACT

Background: Sepsis is a leading cause of morbidity, mortality, and health care costs worldwide.

Methods: We conducted a multi-center, prospective cohort study evaluating the yield of blood cultures drawn before and after empiric antimicrobial administration among adults presenting to the emergency department with severe manifestations of sepsis (ClinicalTrials.gov: NCT01867905). Enrolled patients who had the requisite blood cultures drawn were followed for 90 days. We explored the independent association between blood culture positivity and its time to positivity in relation to 90-day mortality.

Findings: 325 participants were enrolled; 90-day mortality among the 315 subjects followed-up was 25·4% (80/315). Mortality was associated with age (mean age in those who died was 72·5 ±15·8 vs. 62·9 ±17·7 years among survivors, p<0·0001), greater Charlson Comorbidity Index (2 (IQR 1,3) vs. 1 (IQR 0,3), p=0·008), dementia (13/80 (16·2%) vs. 18/235 (7·7%), p=0·03), cancer (27/80 (33·8%) vs. 47/235 (20·0%), p=0·015), positive qSOFA score (57/80 (71·2%) vs. 129/235 (54·9%), p=0·009), and normal white blood cell counts (25/80 (31·2%) vs. 42/235 (17·9%), p=0·02). The presence of bacteremia, persistent bacteremia after antimicrobial infusion, and shorter time to blood culture positivity were not associated with mortality. Neither the source of infection nor pathogen affected mortality.

Interpretation: Although severe sepsis is an inflammatory condition triggered by infection, its 90-day survival is not influenced by blood culture positivity nor its time to positivity.

Funding: Vancouver Coastal Health; St-Paul’s Hospital Foundation Emergency Department Support Fund; the Fonds de Recherche Santé – Québec (CPY); Intramural Research Program of the NIH, Clinical Center (AL); the Maricopa Medical Foundation

Key Words: “sepsis”, “bacteremia”, “bloodstream infection, “mortality”, “survival”
INTRODUCTION

Sepsis is a leading cause of hospitalizations, morbidity, mortality, and health care costs worldwide.\textsuperscript{1} The annual incident cases of sepsis worldwide is estimated at 48·9 million, resulting in 11·0 million sepsis-related deaths.\textsuperscript{2} Despite its significant global burden, the underlying pathophysiology of sepsis remains poorly understood.

Sepsis is recognized as a dysregulated host inflammatory response to systemic infection whose management rests upon timely diagnosis, early administration of antimicrobial therapy, aggressive resuscitation, and supportive care.\textsuperscript{1,3,4} However, despite the institution of sepsis treatment bundles aiming for rapid recognition and early administration of empiric antimicrobial therapy, mortality rates for severe manifestations of sepsis continue to range between 20 and 30\%\textsuperscript{5,6}. Reported risk factors for death include increasing age, immune dysfunction, the source of infection, delays in or receipt of non-effective antimicrobials, and associated organ injury.\textsuperscript{7-13} The time interval between specimen collection and detection of bacterial growth by automated blood culture systems correlates with measures of bacterial inoculum\textsuperscript{14} and is automatically captured in laboratory information systems. There is a relative paucity of data regarding the association between blood culture positivity as well as its time to positivity in relation to 90-day mortality rates among patients with severe manifestations of sepsis.

The initial results of the FABLED diagnostic study (eFfect of Antimicrobial administration on BlOod culture positivity in patients with severe manifestations of sepsis in the Emergency Department), have been reported.\textsuperscript{15} In this pre-specified analysis, we present the FABLED cohort mortality data. We hypothesized that, among patients presenting to the emergency department (ED) with severe manifestations of sepsis, blood culture positivity and a shorter
time to blood culture positivity would be associated with an increased risk of death at 90 days.

METHODS

Study Design

We conducted a multi-center, diagnostic study evaluating the yield of blood cultures drawn prior to and after administration of empiric antimicrobials among adults presenting with severe manifestations of sepsis. Patients were recruited in one of seven EDs across Canada and the United States between November 2013 and September 2018. Details regarding the design, inclusion, exclusion criteria, and diagnostic procedures of the FABLED study have been described. In short, adults ≥18 years of age presenting to the ED with evidence of a systemic inflammatory response syndrome and a presumed or confirmed source of infection were considered for inclusion. Patients required a marker of severity, including either a systolic blood pressure < 90 mmHg or a serum lactate ≥4 mmol/L for enrollment. Enrolled patients who had blood cultures drawn before and after the start of empiric antimicrobial therapy were followed to establish a prospective cohort. The study was approved by the research ethics board at each recruiting center.

Outcomes

The primary outcome of this study was 90-day mortality measured from the time of presentation to the ED. Survival was ascertained using proof of being alive at least 90 days after study recruitment as documented by a hospital or clinic visit in electronic medical records. For those in whom this proof was not available, date of death was confirmed using death certificates obtained from patient medical charts. Subjects for whom we could not unequivocally determine whether they were alive or dead were deemed lost to follow-up.
Covariates of interest included the presence of positive blood cultures before or after initiation of empiric antimicrobial therapy, the time to blood culture positivity, the source of infection, and the causative organism. Blood cultures growing contaminant organisms, defined as low-virulence skin flora within a single set of blood cultures and confirmed as such by infectious diseases and medical microbiology specialists (MPC, CPY), were considered negative. Blood cultures were considered concordant when the growth of the same organism(s) was documented in blood cultures obtained both prior to and after receipt of empiric antimicrobials. In the setting of a polymicrobial infection, all non-contaminant organisms detected in the pre-antimicrobial cultures must have been present in the post-antimicrobial blood cultures to have been considered concordant. Time to positivity of blood cultures was defined as the interval, in hours, between the time of blood culture draw to the time of microbial detection in the automated blood culture system. The blood culture platforms used in the participating institutions and their characteristics have been described. For the purposes of these analyses, the pathogens recovered were categorized as gram positive, gram negative, polymicrobial or fungal species.

**Sample Size**

Sample size calculations for the FABLED diagnostic study yielded a target 328 subjects. Assuming a 10% loss to follow-up with a conservative 35% bacteremia rate per sepsis literature and sepsis epidemiology, we estimated that 103 bacteremic and 190 culture-negative subjects would have 90-day outcome data. Opting for a conservative predicted mortality among patients with severe sepsis, we postulated that 20% of bacteremic study subjects would die within the follow-up period. Since a 5-10% difference in mortality had been reported between persons with culture-positive vs. culture-negative sepsis, we based our estimates on a 5% between-group mortality difference with a 10% standard
deviation. Thus, assuming a sample size of 103 subjects per group (i.e. ≥103 culture-positive subjects), 20% mortality with a 5% between-group difference and 10% standard deviation, and a two-sided alpha error rate of 5%, our follow-up cohort would provide 95% power to detect a difference in mortality between culture-positive and culture-negative subjects.

Statistical Analyses

All analyses were performed using SAS version 9.4 (SAS Institute, Cary, North Carolina, USA). A two-tailed p-value <0.05 was considered significant for all analyses. Patients in whom 90-day mortality data could not be unequivocally determined were deemed lost to follow-up and excluded from the analyses (3% of cohort). All remaining subjects had complete exposure and outcome data ascertainment.

Participants were classified per 90-day mortality status. Categorical variables were analysed as proportions; normally distributed continuous variables using means and standard deviations, while other continuous data were represented with medians and corresponding inter-quartile ranges (IQR). Between group comparisons were made using Fisher’s Exact test for categorical data, t-tests for normally distributed continuous data, and Wilcoxon tests for non-normal data.

Univariate analyses were performed to evaluate for significant differences in microbiologic data between those who did and did not survive to 90 days. Results were presented as odds ratios with their corresponding 95% confidence intervals (95% CI) and p-values. Of note, the relationship between time to positivity and mortality was adjusted for incubation system in the otherwise univariate analyses, per data suggesting a significant difference in the speed of microbial detection per platform. These adjustments were done using a simple logistic regression model with p-values generated by the Wald chi square test.
Logistic regression models were designed to evaluate different positive blood culture characteristics, accounting for potential confounders, effect modifiers, and known predictors of mortality. Characteristics included the presence of a bloodstream infection prior to or after initiation of empiric antimicrobials, concordant blood cultures, and time to positivity among bacteremic patients. Time to positivity was chosen as the primary marker of interest because having a greater bacterial inoculum should result in a higher bacterial count in the blood collected, and thus a shorter time before a critical mass is detected and identified as “positive” by the automated blood culture platform.

The pathogen recovered was analysed categorically without reference parameterization; categories were limited to gram-positive, gram-negative, and polymicrobial. The growth of *Candida* species in only two patients, both of whom survived, precluded the use of this microbial category in a logistic regression model. Source of infection was also analysed categorically without reference parameterization; categories included gastrointestinal, genitourinary, respiratory, skin and soft tissue, other, and unknown source of infection. Evaluating the scatterplot depicting the relationship between time to positivity and mortality suggested that the optimal way to use this continuous variable was in a linear fashion. Because age, the presence of comorbidities, sex, and quick Sequential Organ Failure Assessment (qSOFA) score upon presentation have been associated with sepsis-related mortality in the literature, patient age, sex, Charlson Comorbidity Index, and positive qSOFA score (defined as a qSOFA score of ≥2 per clinical practice) were empirically included in the multivariate models. Since patient baseline characteristics revealed a significant between-group difference in abnormal white blood cell count and malignancy, these two variables were also entered in the models. Finally, although the proportion of patients with dementia was different between groups, it was highly associated with both age and Charlson Comorbidity Index, thus not retained for the models. Multi-collinearity within
the models was assessed. After computing the models, diagnostics were run to evaluate model fit and determine whether the residual data points were normally distributed. None of the models were over-fitted. Results were presented as odds ratios with 95% CIs in the form of a forest plot for ease of visual interpretation.

Role of the Funding Source

The funding bodies had no input as to the study design; collection, analysis, or collection of data, writing of this report nor the decision to submit the paper for publication.

Patient Consent Statement

Written informed consent to participate in the FABLED study was obtained from all patients, or their surrogate decision makers, as previously described.16

RESULTS

The FABLED diagnostic study enrolled 330 participants, five of which were excluded from the cohort study for lack of post-antimicrobial blood cultures and 10 were lost to follow-up. In total, 315 participants were retained in this cohort study (Figure 1). The overall 90-day mortality rate was 25·4%.

Baseline patient characteristics are presented in Table 1. The mean age of participants was 65·3 (± 17·7) years, 199 (63·4%) were male, and the median Charlson Comorbidity Index was 1 (IQR 1, 3). In the ED, 186 (59%) had a positive qSOFA score, 196 (62·2%) had a lactate measured ≥4.0mmol/L, 178 (56·5%) had a systolic blood pressure <90mmHg, 40 (12·7%) had respiratory failure, and 49 (15·6%) required vasopressor support. Of the 102 patients with positive blood cultures prior to receiving antimicrobial therapy, 43 (42·1%) were infected with a gram-positive organism, 45 (44·1%) with a gram-negative organism, 12
(11·8%) had a polymicrobial infection, and 2 (2·0%) had a bloodstream infection with a *Candida* species.

When comparing subjects who died with those who survived to 90-days, patients who died were older (mean age 72·5 ±15·8 vs. 62·9 ±17·7 years, p<0·01), had higher Charlson Comorbidity Index (2 (IQR 1,3) vs. 1 (IQR 0,3), p<0·01), were more likely to have dementia (13/80 (16·2%) vs. 18/235 (7·7%), p=0·03), a malignancy (27/80 (33·8%) vs. 47/235 (20·0%), p=0·02), a positive qSOFA score (57/80 (71·2%) vs. 129/235 (54·9%), p<0·01), and fewer had abnormal white blood cell (WBC) counts (55/80 (68·8%) vs. 193/235 (82·1%), p=0·02). There was no significant difference in lactate, systolic blood pressure, respiratory failure or vasopressor requirement in the ED between those who died and those who survived.

Blood culture results were assessed several ways, including the presence of bacteremia, persistent bacteremia, and time to positivity among bacteremic patients. None of these markers were associated with mortality on univariate analysis (Table 2). Likewise, the source of infection and the causative organism were not associated with mortality (Table 2). When entering each of the individual blood culture characteristics in a multivariate model adjusting for age, male sex, Charlson Comorbidity Index, underlying malignancy, positive qSOFA score, and abnormal WBC count, the lack of association persisted (Table 3). When studied alone or adjusting for any of the blood culture results, the impact of a positive qSOFA score on the risk of mortality remained significant and unchanged (OR 2·03 (95% CI 1·17, 3·51)), as did increasing age (per year increase in OR 1·03 (95% CI 1·02, 1·05)) and the presence of an abnormal WBC count (OR 0·50 (95% CI 0·27, 0·92)).
DISCUSSION

Our results demonstrate that several patient characteristics are associated with an increased risk of 90-day mortality among patients with severe manifestations of sepsis, including a positive qSOFA score. This finding is concordant with the published literature and supports the use of the qSOFA score in the emergency department to classify individuals at higher risk of mortality. However, we were unable to demonstrate an association between blood culture positivity or its time to positivity in relation to 90-day mortality on univariate and multivariate analyses. While essential for antimicrobial selection and stewardship practices, our data suggest that blood culture results in isolation cannot be used to predict mortality in this patient population.

We assessed blood culture results in three orthogonal and complementary ways. First, we evaluated whether the presence of bacteremia, before administration of empiric antimicrobials, after initiation of antimicrobial therapy, or at any time, was associated with an increased risk of 90-day mortality. In univariate and multivariate analyses, controlling for confounders and effect modifiers that might mask its effect, bacteremia was not associated with an increased risk of mortality at 90 days. Second, we assessed whether the occurrence of bacteremia both prior to and following initiation of antimicrobial therapy might represent greater disease severity and thus have an impact on the observed 90-day mortality. Again, in both univariate and multivariate analyses, this was not the case. We then sought to establish whether a marker of the extent of infectious inoculum, i.e. the rapidity of microbial growth in blood culture media, would modify the 90-day mortality risk. Measuring the effect of time to positivity, amongst blood cultures positive either before or even after administration of antimicrobials, had no effect on the risk of death.

Other recent cohorts have sought to assess whether the presumed source site of infection or specific pathogen recovered might influence a septic patient’s risk of mortality. A
recent meta-analysis highlights that the source of infection is unlikely to be a significant
determinant of mortality in this population\textsuperscript{25}. The 10-year prospective administrative cohort
by Zahar \textit{et al}, which included more than 3,500 patients with severe sepsis, reported that
pathogen species and the site of infection did not influence mortality\textsuperscript{26}. Our study
corroborates these findings as neither the source of infection nor causative pathogen were
associated with a difference in 90-day mortality. Once patients develop severe manifestations
of sepsis, it is possible that microbial factors become relatively less important to determining
clinical outcomes as a dysregulated host response becomes responsible for disease
manifestations. The prognostic significance of microbial factors in earlier stages of sepsis
may be different than in our study population.

Our study has several strengths. First, we recruited a critically ill group of patients
(32.6\% had a positive blood culture, 25\% died within 90 days) and had excellent study
retention (97\% of the study population). Furthermore, we performed a pragmatic study
reflecting standards of practice in ED and microbiology laboratories across seven centers in
Canada and the United States, increasing the generalizability to the results.

A potential limitation of our study is that the quantity of blood drawn for culture
varied between study sites due to heterogeneous laboratory practices and differences in study
protocol per local research ethics board specifications. However, the total blood volume
cultured from subjects at all participating centers before and after antimicrobial
administration was \( \geq 60 \) mL, which should suffice for pathogen recovery\textsuperscript{27}. Furthermore, our
sample size was insufficiently large to observe significant differences from certain bacterial
species or different antimicrobial therapies on the risk of mortality. For example, it is possible
that a larger cohort study would have found a statistically significant relationship between
organism and 90-day risk of mortality, as patients with positive follow-up blood cultures with
Gram-negative organisms are known to be at increased risk of poor outcomes\textsuperscript{28}. 
In conclusion, neither blood culture positivity nor its time to positivity was associated with 90-day mortality among adults presenting to the ED with severe manifestations of sepsis. These findings support the ongoing use and research into bundles of care aimed at treating both the infectious and inflammatory aspects of sepsis, since blood culture results alone do not appear to be a significant driver of mortality in this patient population.
ACKNOWLEDGEMENTS

FUNDING: This work was supported by Vancouver Coastal Health; St-Paul’s Hospital Foundation Emergency Department Support Fund; the Fonds de Recherche Santé – Québec to CPY; Intramural Research Program of the NIH, Clinical Center (AL); and the Maricopa Medical Foundation.

Potential Conflicts of Interest: The authors have no relevant conflicts of interest to declare. MPC reports receiving personal fees from GEn1E Lifesciences (as a member of the scientific advisory board) and personal fees from nplex biosciences (as a member of the scientific advisory board), outside the submitted work. He is also the cofounder of Kanvas Biosciences.
REFERENCES

1. Cohen J, Vincent JL, Adhikari NK, et al. Sepsis: a roadmap for future research. 
   *Lancet Infect Dis.* 2015;15(5):581-614.
2. Rudd KE, Johnson SC, Agesa KM, et al. Global, regional, and national sepsis 
   incidence and mortality, 1990-2017: analysis for the Global Burden of Disease Study. 
   *Lancet.* 2020;395(10219):200-211.
3. Rhodes A, Evans LE, Alhazzani W, et al. Surviving Sepsis Campaign: International 
   Guidelines for Management of Sepsis and Septic Shock: 2016. *Crit Care Med.* 
   2017;45(3):486-552.
4. Singer M, Deutschman CS, Seymour CW, et al. The Third International Consensus 
   Definitions for Sepsis and Septic Shock (Sepsis-3). *JAMA.* 2016;315(8):801-810.
5. Fleischmann C, Scherag A, Adhikari NK, et al. Assessment of Global Incidence and 
   Mortality of Hospital-treated Sepsis. Current Estimates and Limitations. *Am J Respir 
   Crit Care Med.* 2016;193(3):259-272.
6. Stevenson EK, Rubenstein AR, Radin GT, Wiener RS, Walkey AJ. Two decades of 
   mortality trends among patients with severe sepsis: a comparative meta-analysis*. 
   *Crit Care Med.* 2014;42(3):625-631.
7. Seymour CW, Gesten F, Prescott HC, et al. Time to Treatment and Mortality during 
   Mandated Emergency Care for Sepsis. *N Engl J Med.* 2017;376(23):2235-2244.
8. Liu VX, Fielding-Singh V, Greene JD, et al. The Timing of Early Antibiotics and 
   Hospital Mortality in Sepsis. *Am J Respir Crit Care Med.* 2017;196(7):856-863.
9. 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(6):1589-1596.
10. Sterling SA, Miller WR, Pryor J, Puskarich MA, Jones AE. The Impact of Timing of 
    Antibiotics on Outcomes in Severe Sepsis and Septic Shock: A Systematic Review 
    and Meta-Analysis. *Crit Care Med.* 2015;43(9):1907-1915.
11. Rhee C, Dantes R, Epstein L, et al. Incidence and Trends of Sepsis in US Hospitals 
    Using Clinical vs Claims Data, 2009-2014. *JAMA.* 2017;318(13):1241-1249.
12. Rannikko J, Syrjanen J, Seiskari T, Aittoniemi J, Huttunen R. Sepsis-related mortality 
    in 497 cases with blood culture-positive sepsis in an emergency department. *Int J 
    Infect Dis.* 2017;58:52-57.
13. McCormack D, Ruderman A, Menges W, Kulkarni M, Murano T, Keller SE. 
    Usefulness of the Mortality in Severe Sepsis in the Emergency Department score in 
    an urban tertiary care hospital. *Am J Emerg Med.* 2016;34(6):1117-1120.
14. Haimi-Cohen Y, Vellozzi EM, Rubin LG. Initial concentration of Staphylococcus 
    epidermidis in simulated pediatric blood cultures correlates with time to positive 
    results with the automated, continuously monitored BACTEC blood culture system. *J Clin Microbiol.* 2002;40(3):898-901.
15. Cheng MP, Stenstrom R, Paquette K, et al. Blood Culture Results Before and After 
    Antimicrobial Administration in Patients With Severe Manifestations of Sepsis: A 
    Diagnostic Study. *Ann Intern Med.* 2019;171(8):547-554.
16. Cheng MP, Stenstrom R, Paquette K, et al. Blood Culture Results Before and After 
    Antimicrobial Administration in Patients With Severe Manifestations of Sepsis: A 
    Diagnostic Study. *Ann Intern Med.* 2019.
17. Bone RC, Balk RA, Cerra FB, et al. Definitions for sepsis and organ failure and 
    guidelines for the use of innovative therapies in sepsis. The ACCP/SCCM Consensus 
    Conference Committee. American College of Chest Physicians/Society of Critical 
    Care Medicine. *Chest.* 1992;101(6):1644-1655.
18. Baron EJ, M. P. Weinstein, W. M. Dunne, Jr., P. Yagupsky, D. F. Welch, and D. M. Wilson. 2005. Cumitech 1C, Blood Cultures IV. Coordinating ed., E. J. Baron. ASM Press, Washington, D.C.

19. Kumar A, Zarychanski R, Light B, et al. Early combination antibiotic therapy yields improved survival compared with monotherapy in septic shock: a propensity-matched analysis. Crit Care Med. 2010;38(9):1773-1785.

20. Pro CI, Yealy DM, Kellum JA, et al. A randomized trial of protocol-based care for early septic shock. N Engl J Med. 2014;370(18):1683-1693.

21. Gupta S, Sakhija A, Kumar G, McGrath E, Nanchal RS, Kashani KB. Culture-Negative Severe Sepsis: Nationwide Trends and Outcomes. Chest. 2016;150(6):1251-1259.

22. Phua J, Ngerng W, See K, et al. Characteristics and outcomes of culture-negative versus culture-positive severe sepsis. Crit Care. 2013;17(5):R202.

23. Butler-Laporte G, Yansouni CP, Paquette K, et al. Real-world Time to Positivity of 2 Widely Used Commercial Blood Culture Systems in Patients With Severe Manifestations of Sepsis: An Analysis of the FABLED Study. Open Forum Infect Dis. 2020;7(9):ofaa371.

24. Jiang J, Yang J, Mei J, Jin Y, Lu Y. Head-to-head comparison of qSOFA and SIRS criteria in predicting the mortality of infected patients in the emergency department: a meta-analysis. Scand J Trauma Resusc Emerg Med. 2018;26(1):56.

25. Motzkus CA, Luckmann R. Does Infection Site Matter? A Systematic Review of Infection Site Mortality in Sepsis. J Intensive Care Med. 2017;32(8):473-479.

26. Zahar JR, Timsit JF, Garrouste-Orgeas M, et al. Outcomes in severe sepsis and patients with septic shock: pathogen species and infection sites are not associated with mortality. Crit Care Med. 2011;39(8):1886-1895.

27. Baron EJ, Scott JD, Tompkins LS. Prolonged incubation and extensive subculturing do not increase recovery of clinically significant microorganisms from standard automated blood cultures. Clin Infect Dis. 2005;41(11):1677-1680.

28. Maskarinec SA, Park LP, Ruffin F, et al. Positive follow-up blood cultures identify high mortality risk among patients with Gram-negative bacteraemia. Clin Microbiol Infect. 2020;26(7):904-910.
### Table 1 – Patient Characteristics per 90-Day Mortality

| Characteristic                              | Survived n= 235 (100%) | Died n= 80 (100%) | All n= 315 (100%) |
|---------------------------------------------|-------------------------|-------------------|-------------------|
| Mean age in years (SD)\(^a\)                | 62.9 (17.7)             | 72.5 (15.8)       | 65.3 (17.7)       |
| Male sex                                    | 154 (65.8)              | 45 (56.2)         | 199 (63.4)        |
| Comorbidities                               |                         |                   |                   |
| Chronic kidney disease                      | 19 (8.1)                | 5 (6.2)           | 24 (7.6)          |
| Chronic pulmonary disease                   | 47 (20.0)               | 18 (22.5)         | 65 (20.6)         |
| Cirrhosis                                   | 4 (1.7)                 | 2 (2.5)           | 6 (1.9)           |
| Congestive heart failure                    | 26 (11.1)               | 10 (12.5)         | 36 (11.4)         |
| Dementia \(^a\)                             | 18 (7.7)                | 13 (16.2)         | 31 (9.8)          |
| Diabetes                                    | 60 (25.5)               | 25 (31.2)         | 85 (27.0)         |
| HIV or AIDS                                 | 15 (6.4)                | 1 (1.2)           | 16 (5.1)          |
| Malignancy \(^a\)                          | 47 (20.0)               | 27 (33.8)         | 74 (23.5)         |
| Charlson Comorbidity Index, median (IQR) \(^b\) | 1 (0.3)                    | 2 (1.3)            | 1 (1.3)            |
| qSOFA Score \(^a\)                          |                         |                   |                   |
| 0                                           | 20 (8.5)                | 10 (12.5)         | 30 (9.5)          |
| 1                                           | 86 (36.6)               | 13 (16.2)         | 99 (31.4)         |
| 2                                           | 90 (38.3)               | 40 (50)           | 140 (41.3)        |
| 3                                           | 39 (16.6)               | 17 (21.2)         | 56 (18.3)         |
| qSOFA Positive \(^a\)                       | 129 (54.9)              | 57 (71.2)         | 186 (59.0)        |
| Initial characteristics in the emergency department |                         |                   |                   |
| Heart Rate > 90 beats per minute            | 196 (83.4)              | 63 (78.8)         | 259 (82.2)        |
| Temperature >38°C or <36°C                   | 144 (61.3)              | 46 (57.5)         | 190 (60.3)        |
| White Blood Cell >12 or <4, x1,000/mL \(^a\) | 193 (82.1)              | 55 (68.8)         | 248 (78.7)        |
| Lactate ≥4.0 mmol/L                         | 145 (61.7)              | 51 (63.8)         | 196 (62.2)        |
| Systolic BP <90mmHg                         | 132 (56.2)              | 46 (57.5)         | 178 (56.5)        |
| Respiratory failure                         | 30 (12.8)               | 10 (12.5)         | 40 (12.7)         |
| Vasopressor requirement                     | 35 (14.9)               | 14 (17.5)         | 49 (15.6)         |
| Source of Infection                         |                         |                   |                   |
| Gastrointestinal                            | 40 (17.0)               | 13 (16.2)         | 53 (16.8)         |
| Genitourinary                               | 44 (18.7)               | 12 (15.0)         | 56 (17.8)         |
| Respiratory                                 | 71 (31.2)               | 33 (41.2)         | 104 (33.0)        |
| Skin and Soft Tissue                        | 31 (13.2)               | 8 (10.0)          | 39 (12.4)         |
| Other                                       | 11 (4.7)                | 4 (5.0)           | 15 (4.8)          |
| Unknown                                     | 38 (16.2)               | 10 (12.5)         | 48 (15.2)         |
| Pathogen Recovered (n= 102)                 |                         |                   |                   |
| Gram-Positive                               | 38 (47.5)               | 5 (22.7)          | 43 (42.1)         |
| Gram-Negative                               | 31 (38.8)               | 14 (63.4)         | 45 (44.1)         |
| Polymicrobial                               | 9 (11.2)                | 3 (13.6)          | 12 (11.8)         |
| Candida species                             | 2 (2.5)                 | 0 (0)             | 2 (2.0)           |

\(^a\) Significant differences were observed between groups with regards to: age (p < 0.01), dementia (p=0.03), malignancy (p=0.02), Charlson Comorbidity Index (p<0.01), qSOFA ordinal score (p<0.01), qSOFA positivity defined as a score of qSOFA ≥2 (p<0.01), and abnormal white blood cell count (p=0.02).
Table 2 – Microbial Data and its Relationship to 90-Day Mortality per Univariate Analyses

| Characteristic                                      | Survived n= 235 (100%) | Died n= 80 (100%) | Odd Ratio (95% CI) | p-value |
|-----------------------------------------------------|------------------------|-------------------|--------------------|---------|
| Any Positive Blood Culture\(^1\)                    | 83 (35.3)              | 23 (28.8)         | 0.74 (0.42, 1.28)  | 0.34    |
| Concordant Blood Cultures\(^2\)                    | 44 (18.7)              | 15 (18.8)         | 1.00 (0.52, 1.91)  | 1.0     |
| Pathogen Recovered\(^3\)                           | 80 (34.0)              | 22 (27.8)         | 1.17 (0.99, 1.38)  | 0.06    |
| Source of Infection\(^4\)                          | 235 (100)              | 80 (100)          | 0.97 (0.82, 1.14)  | 0.69    |
| Time to positivity of pre-antimicrobial blood culture, hours, median (IQR) | 14.0 (12, 17)          | 12.0 (10, 17)     | 1.01 (0.93, 1.09)  | 0.86    |
| Time to positivity of post-antimicrobial blood culture, hours, median (IQR) | 17.5 (14, 29)          | 15.5 (11, 23)     | 0.97 (0.92, 1.01)  | 0.16    |

\(^1\)Refers to a positive blood culture either pre or post antimicrobial administration.

\(^2\)Refers to a positive blood culture in which all microbial pathogens recovered from the pre-antimicrobial blood culture are recovered in the post-antimicrobial blood culture.

\(^3\)Refers to the pathogen recovered; gram-positive (n=43), gram-negative (n=45), polymicrobial (n=12), or *Candida* species (n=2).

\(^4\)Refers to the source of infection; gastrointestinal (n=53), genitourinary (n=56), respiratory (n=104), skin and soft tissue (n=39), other (n=15), or unknown (n=48).
| Characteristic                                      | Survived n= 235 | Died n= 80 | Total n=315 | Odd Ratio (95% CI) | p-value |
|----------------------------------------------------|-----------------|------------|-------------|-------------------|--------|
| Positive Blood Culture Prior to Antimicrobials, n (%) | 80 (34.0)       | 22 (27.5)  | 102 (32.4)  | 0.66 (0.37, 1.20)  | 0.18   |
| Positive Blood Culture After Antibiotics, n (%)     | 47 (20.0)       | 16 (20.0)  | 63 (20.0)   | 0.79 (0.40, 1.57)  | 0.51   |
| Any Positive Blood Culture, n (%)                  | 83 (35.3)       | 23 (28.8)  | 106 (33.6)  | 0.65 (0.36, 1.18)  | 0.16   |
| Concordant Blood Cultures, n (%)                   | 44 (18.7)       | 15 (18.8)  | 59 (18.7)   | 0.81 (0.40, 1.63)  | 0.56   |
| Pathogen Recovered                                 | 80 (34.0)       | 22 (27.8)  | 102 (32.3)  | 1.07 (0.89, 1.29)  | 0.51   |
| Source of Infection                                | 235 (100)       | 80 (100)   | 315 (100)   | 0.99 (0.83, 1.17)  | 0.88   |
| Time to positivity of pre-antibiotic blood culture, hours, median (IQR) n=100 | n=79 14.0 (12, 17) | n=21 12.0 (10, 17) | 13.0 (11, 17) | 1.02 (0.93, 1.12) | 0.69 |
| Time to positivity of post-antibiotic blood culture, hours, median (IQR) n=60 | n=44 17.5 (14, 29) | n=16 15.5 (11, 23) | 17.0 (13, 24) | 0.96 (0.91, 1.02) | 0.18 |

*Multivariate model adjusting for age, Charlson Comorbidity Index, the presence of malignancy, a positive qSOFA score, an abnormal WBC count, and male sex.
FIGURES

Figure 1 – Study Flow Diagram
Figure 1 – Study Flow Diagram

Participants were assessed for eligibility (n=3164)

- Participants not eligible because they did not meet study inclusion criteria (n=2722)

Participants eligible (n=442)

- Participants excluded (n=112)
  - Declined to participate: 61
  - Assessed >2 hours after initiation of antimicrobial therapy: 35
  - Did not receive antibiotic therapy in the emergency department: 8
  - Had a hypocoagulable state: 5
  - Died before a repeat blood culture was obtained: 3

Participants enrolled into the study (n=330)

- Did not have repeat blood cultures drawn within 4 hours of initiation of antimicrobials (n=5)

Participants included in the diagnostic study who formed the basis for the follow-up cohort (n=325)

- Lost to follow-up (n=10)

Participants included in the mortality analyses (n=315)