

A Multicenter Evaluation of Blood Culture Practices, Contamination Rates, and the Distribution of Causative Bacteria

Mustafa Altindis,^{1*} Mehmet Koroglu,¹ Tayfur Demiray,² Tuba Dal,³ Mehmet Ozdemir,⁴ Ahmet Zeki Sengil,⁵ Ali Riza Atasoy,¹ Metin Doğan,⁴ Aysegul Copur Cicek,⁶ Gulfem Ece,⁷ Selcuk Kaya,⁸ Meryem Iraz,⁹ Bilge Sumbul Gultepe,⁹ Hakan Temiz,¹⁰ Idris Kandemir,¹¹ Sebahat Aksaray,¹² Yeliz Cetinkol,¹³ Idris Sahin,¹⁴ Huseyin Guducuoglu,¹⁵ Abdullah Kilic,¹⁶ Esra Kocoglu,¹⁷ Baris Gulhan,¹⁸ and Oguz Karabay¹⁹

¹Department of Clinical Microbiology, Faculty of Medicine, Sakarya University, Sakarya, Turkey

²Department of Clinical Microbiology, Training and Research Hospital, Sakarya University, Sakarya, Turkey

³Department of Clinical Microbiology, School of Medicine, Yildirim Beyazit University, Ankara, Turkey

⁴Department of Clinical Microbiology, Meram Medical Faculty Hospital, Necmettin Erbakan University, Konya, Turkey

⁵Department of Medical Microbiology, Medical Faculty, Medipol University, Istanbul, Turkey

⁶Department of Clinical Microbiology, School of Medicine, Recep Tayyip Erdogan University, Rize, Turkey

⁷Department of Clinical Microbiology, School of Medicine, Izmir University, Izmir, Turkey

⁸Department of Clinical Microbiology, School of Medicine, Izmir Katip Celebi University, Izmir, Turkey

⁹Department of Clinical Microbiology, School of Medicine, Bezmi Alem University, Istanbul, Turkey

¹⁰Department of Clinical Microbiology, Diyarbakir Training and Research Hospital, Diyarbakir, Turkey

¹¹Department of Clinical Microbiology, School of Medicine, Dicle University, Diyarbakir, Turkey

¹²Department of Clinical Microbiology, Haydarpasa Numune Hospital, Istanbul, Turkey

¹³Department of Clinical Microbiology, School of Medicine, Ordu University, Ordu, Turkey

¹⁴Department of Clinical Microbiology, School of Medicine, Duzce University, Duzce, Turkey

¹⁵Department of Clinical Microbiology, School of Medicine, Yuzuncuyl University, Van, Turkey

¹⁶Department of Clinical Microbiology, School of Medicine, Gulhane Military Medical School, Ankara, Turkey

¹⁷Department of Clinical Microbiology, School of Medicine, Abant Izzet Baysal University, Bolu, Turkey

¹⁸Department of Clinical Microbiology, School of Medicine, Erzincan University, Erzincan, Turkey

¹⁹Department of Infection Diseases, School of Medicine, Sakarya University, Sakarya, Turkey

*Corresponding author: Mustafa Altindis, Department of Clinical Microbiology, Faculty of Medicine, Sakarya University, Sakarya, Turkey. Tel: +90-2642957277, Fax: +90-2642956629, E-mail: maltindis@gmail.com

Received 2015 May 13; Revised 2015 September 3; Accepted 2015 September 22.

Abstract

Background: The prognostic value of blood culture testing in the diagnosis of bacteremia is limited by contamination.

Objectives: In this multicenter study, the aim was to evaluate the contamination rates of blood cultures as well as the parameters that affect the culture results.

Materials and Methods: Sample collection practices and culture data obtained from 16 university/research hospitals were retrospectively evaluated. A total of 214,340 blood samples from 43,254 patients admitted to the centers in 2013 were included in this study. The blood culture results were evaluated based on the three phases of laboratory testing: the pre-analytic, the analytic, and the post-analytic phase.

Results: Blood samples were obtained from the patients through either the peripheral venous route (64%) or an intravascular catheter (36%). Povidone-iodine (60%) or alcohol (40%) was applied to disinfect the skin. Of the 16 centers, 62.5% have no dedicated phlebotomy team, 68.7% employed a blood culture system, 86.7% conducted additional studies with pediatric bottles, and 43.7% with anaerobic bottles. One center maintained a blood culture quality control study. The average growth rate in the bottles of blood cultures during the defined period (1259 - 26,400/year) was 32.3%. Of the growing microorganisms, 67% were causative agents, while 33% were contaminants. The contamination rates of the centers ranged from 1% to 17%. The average growth time for the causative bacteria was 21.4 hours, while it was 36.3 hours for the contaminant bacteria. The most commonly isolated pathogens were *Escherichia coli* (22.45%) and coagulase-negative staphylococci (CoNS) (20.11%). Further, the most frequently identified contaminant bacteria were CoNS (44.04%).

Conclusions: The high contamination rates were remarkable in this study. We suggest that the hospitals' staff should be better trained in blood sample collection and processing. Sterile glove usage, alcohol usage for disinfection, the presence of a phlebotomy team, and quality control studies may all contribute to decreasing the contamination rates. Health policy makers should therefore provide the necessary financial support to obtain the required materials and equipment.

Keywords: Blood Specimen Collection, Phlebotomy, Blood-Borne Pathogens, Bacteriological Techniques

1. Background

Bacteremia is a common cause of morbidity and mortality in hospitalized patients (1). The early and accurate identification of the causative organism is therefore necessary for patient survival. The blood culture test is considered to be the “gold standard” in the diagnosis and treatment of bacteremia. However, the prognostic value of blood culture testing is limited by contamination (2, 3). A blood culture contaminant is defined as a microorganism isolated from a blood culture that was introduced into the culture during either specimen collection or processing and that was not pathogenic for the patient from whom the blood was collected (2).

The most common contaminant microorganisms are coagulase-negative staphylococci and other skin flora species such as *Viridans streptococci*, *Corynebacterium* species other than *C. jeikeium*, *Bacillus* species, and *Propionibacterium acnes* (4). The Standards of the American Society for Microbiology and the Clinical and Laboratory Standards Institute (CLSI) state that acceptable contamination rates should be no higher than 2 to 3% (5, 6). Patients with contaminated blood cultures often receive unnecessary antibiotics, and additional laboratory tests are needed to determine the cause of the positive blood culture test. Contaminated blood cultures also lead to an increased length of hospital stay, increased costs, increased work load, and the unnecessary removal of central intravenous lines (7, 8).

High quality blood culture results are dependent on evaluation during the three phases of laboratory testing: the pre-analytic, the analytic, and the post-analytic phase. Recently, the use of sensitive automated blood culture systems with rich culture media has led to increased contamination rates (2, 9, 10).

2. Objectives

In this multicenter study, we aimed to evaluate the contamination rates of blood cultures, as well as the primary parameters affecting the culture results, throughout the entire process from the collection of the blood culture to the interpretation of the results in different tertiary care hospitals in Turkey.

3. Materials and Methods

In this study, sample collection practices and culture data obtained from 16 university/research hospitals were retrospectively analyzed in 2013. A total of 214,340 blood samples collected from 43,254 patients who were admitted to the centers in 2013 were included in the analysis. The study centers were: Sakarya University Training and Research Hospital, Sakarya; Necmettin Erbakan University Meram Medical Faculty Hospital, Konya; Medipol University Medical Faculty, Istanbul; Recep Tayyip Erdogan University, Rize; Izmir University Medical Faculty, Izmir; Izmir Katip Celebi University Medical Faculty, Izmir; Bezmi Alem University Medical Faculty, Istanbul; Diyarbakir Training and Research Hospital, Diyarbakir; Dicle University Medical Faculty, Diyarbakir; Haydarpasa Numune Hospital, Istanbul; Ordu University Medical Fac-

ulty, Ordu; Duzce University Medical Faculty, Duzce; Yuzuncuyil University Medical Faculty, Van; GATA Medical Faculty, Ankara; Abant Izzet Baysal University Medical Faculty, Bolu; and Erzincan University Medical Faculty, Erzincan. All necessary forms, including the daily practices of the centers, were completed by each individual center and then collected at Sakarya University Training and Research Hospital, Sakarya (Figure 1).

The blood culture bottles were incubated in Bactec™ BD 9120 and 9240 (Becton Dickinson, MD, USA), BacT/ALERT (bioMérieux, Durham, NC, USA), and VERSATEK blood culture (TREK Diagnostic Systems, Cleveland, Ohio) systems at 37°C for 7 - 10 days. After growth, the culture samples were inoculated onto 5% sheep blood agar (Oxoid Ltd., Basingstoke, UK) and the plate was incubated at 36.8°C for 18 - 24 hours. Isolate identification was performed using the BD Phoenix™ 100 (Becton Dickinson, MD, USA) and VITEK 2 (bioMérieux, Marcy l'Etoile, France) fully automated microbiology systems and conventional methods. The blood culture results were evaluated based on the three phases of laboratory testing: the pre-analytic, the analytic, and the post-analytic phase. The evaluated parameters included patient variables, specimen variables, collection, handling, and processing in the pre-analytic phase; the performance of selected laboratory tests in the analytic phase; and test reporting variables, recording, reporting, and interpreting in the post-analytic phase (9).

4. Results

4.1. Pre-Analytic Phase Evaluation

The blood samples from the patients were obtained through either the peripheral venous route (64%) or an intravascular catheter (36%). Povidone-iodine (60%) or alcohol (40%) was applied to disinfect the skin prior to blood sampling (Table 1).

Across all the centers, our analysis reveals that 62.5% of them do not have a dedicated phlebotomy team; in 93.7% of them blood is drawn while wearing gloves; 73.3% of them cleanse the bottle stoppers; and the term set is recognized as aerobic bottles obtained from two different arms (80%) or one aerobic bottle plus one anaerobic bottle both obtained from the same arm (20%) (Table 1).

4.2. Analytic Phase Evaluation

We determined that 68.7% of the centers employed the BacT/ALERT (bioMérieux, Durham, NC, USA) blood culture system. Further, 86.7% of the centers conducted additional studies with pediatric bottles, 43.7% with anaerobic bottles, and 66.6% with fungal bottles. All of the laboratories have established critical value reporting, although only one (7.1%) of them maintains a blood culture quality control study.

4.3. Post-Analytic Phase Evaluation

Some 40% of the centers recorded the point of time at

which the relevant device gave the initial growth signal; 80% of the centers carried out Gram staining upon the detection of a signal, while 80% did not perform Gram staining for the bottles with no recorded signal. As a result of the assessments, the average growth rate in blood culture bottles sent for testing during the defined period (1259 - 26400/year) was calculated to be 32.3%. Out of the growing microorganisms, 67% were described as causative agents, while 33% were referred to as contaminants. The contamination rates reported by the centers ranged from 1% to 17%. The average growth time for the bacteria that were accepted as causative agents was 21.4 hours, while it took an average of 36.3 hours for contaminant bacteria to grow (Table 1).

The most common pathogens that grew in the blood cultures were identified as, in decreasing order, *Escherichia coli*

(22.45%), coagulase-negative Staphylococci CoNS (20.11%), *Enterococci* spp. (9.41%), *Klebsiella* spp. (9.18%), *Staphylococcus aureus* (7.87%), *Pseudomonas aeruginosa* (7.46%), *Acinetobacter baumannii* (6.44%), methicillin-resistant coagulase-negative Staphylococci (5.88%), and other members of the *Enterobacteriaceae* family (5.20%) (Table 2). The most frequently identified contaminant bacteria were CoNS (44.04%), *Diphtheroid bacilli* (32.13%), *Streptococcus* spp. (6.81%), and others (17.03%).

The opinion of the physician, the number of positive blood culture bottles, and any inflammation marker levels (such as white blood cell count, procalcitonin, and CRP) were all considered when determining whether a particular bacterial growth represented a causative agent or a contamination in all of the centers.



Figure 1. Location of Centers Participating in the Study

Table 1. The Ratios Related to the Collection and Processing of Blood Cultures in 16 University or Research and Training Hospitals in Turkey in 2013

Variables	Values ^a
Infection/contamination rates in isolated microorganisms	
Contamination	33
Causative agent	67
Skin disinfection	
Povidone-iodine	60
Alcohol	40
Staff collecting BC	
Nurses	57.7
Physicians	30.7
Medical interns	11.5
Availability of phlebotomy team in centers	37.5

Availability of pediatric bottles in centers	86.7
Availability of anaerobic bottles in centers	66.6
Average growth rate in the bottles	32.3
The average growth time for causative agents, h	21.4
The average growth time for contaminant agents, h	36.3
Route of BC collection	
Intravenous catheter	36
Peripheral venipuncture	64
Hospital classification	
University hospital	56.3
Training and research hospital	43.7
Hospitals with ≥ 500 beds	56.25
Report of growing signal time to clinicians	40
Overall rate of glove usage in the centers	93.75
Number of bottles for diagnosis (≥ 2)	77.8
Using conventional identification methods	28.6
Quality control application	7.1
Sample rejection criteria	
Insufficient blood samples	30.8
Registration errors	69.2
Fungal blood culture assessment	66.7

Abbreviation: BC, Blood cultures.

^aData are presented as percentage.

Table 2. Distribution of Species (%) Isolated From Blood Cultures in 16 Different Hospitals in Turkey in 2013

Microorganisms	Values ^a
<i>Escherichia coli</i>	22.45
CoNS	20.11
MRCoNS	5.88
<i>Enterococcus spp.</i>	9.41
<i>Klebsiella spp.</i>	9.18
<i>Staphylococcus aureus</i>	7.87
<i>Pseudomonas aeruginosa</i>	7.46
<i>Acinetobacter baumannii</i>	6.44
Other members of <i>Enterobacteriaceae</i>	5.2

Abbreviations: BC, Blood cultures; CoNS, Coagulase-negative staphylococci; MRCoNS, Methicillin-resistant coagulase-negative staphylococci.

^aData are presented as percentage.

5. Discussion

Bloodstream infections are a significant cause of mortality and morbidity in any hospital setting. The reported mortality rate worldwide due to bloodstream infections is between 30% and 55% (11-14). Increasing the reliability of blood culture results and reducing contamination rates are both related to the pre-analytic, analytic, and post-analytic phases of laboratory testing (15-17). Of the three phases, the most errors occur during the pre-analytic phase, and most such errors are related to specimen collection, specimen handling, and patient variables (18).

According to the literature, the collection of specimens from intravenous catheters is associated with higher blood culture contamination rates (19). Using a direct venous puncture to a peripheral vein is therefore recommended for obtaining higher specificity and positive predictive power (19). In a systematic review, venous puncture was suggested as the most appropriate method to decrease blood culture contamination (4). In the current study, of all the blood culture samples, 64% were collected from peripheral venous blood, while 36% were

collected from intravascular catheters. The collection of specimens from intravenous catheters may hence be the reason for our high contamination rates.

In the present study, povidone-iodine (60%) and alcohol (40%) were used for skin disinfection prior to blood collection. It has been reported that alcohol is superior to products without alcohol when it comes to skin disinfection prior to blood collection due to alcohol's quick drying time (20, 21). Many researchers have stated that alcohol alone is sufficient, since it is more cost-effective and time-effective than isopropyl alcohol in combination with povidone-iodine (19-21). Our high contamination rates may be related to the reference for using povidone-iodine in the centers. It is suggested to be necessary to wait at least 3 minutes after the application of povidone-iodine for the emergence of an antiseptic effect. The contamination rates may therefore be due to an unwillingness to comply with the waiting period. Mimosz et al. indicated that chlorhexidine reduced the incidence of blood culture contamination more than povidone-iodine. They suggested that skin preparation using alcoholic chlorhexidine was more efficacious in reducing the contamination of blood cultures than skin preparation using aqueous povidone-iodine (22). Based on our findings, it is suggested that the use of alcohol should be increased in our hospital setting.

On the other hand, our study indicated that the ratio of wearing gloves and decontaminating the blood culture bottles prior to use were lower in our centers. Blood culture bottle tops may be nonsterile even if they are covered with a lid, since the sterility of the top varies by manufacturer. Although Bekiris et al. found no correlation between blood culture contamination and the cleaning of culture bottle tops (23), the clinical laboratory standards institute recommends that culture bottle tops be cleaned with 70% isopropyl alcohol (6). Based on our results, it is concluded that routine sterile gloving may decrease blood culture contamination and that cleaning culture bottle tops may also decrease the contamination rates. In the current study, the blood samples were collected by nurses, doctors, and interns. This was necessary because some 62.5% of the centers included in this study had no phlebotomy team. Blood culture contamination is lower when experienced and specialized staff collect the blood samples and so a dedicated phlebotomy team should ideally draw the blood samples for culture (15-19).

Various sensitive blood culture systems and blood culture bottles were used in the Turkish centers. The use of sensitive automated blood culture systems with rich culture media has led to increased contamination rates. In the literature, the most common organisms that indicate a contaminated specimen were CoNS, *Propionibacterium* spp., *Micrococcus* spp., coryneform-type bacilli, *Lactobacillus* spp., *Bacillus* spp., and *Viridans streptococci* (4). The most common contaminant bacteria in the present study were coagulase-negative *Staphylococcus* sp., coryneform-type bacilli, and *Streptococcus* sp., which is similar to findings in the literature. In addition, only one of the

units included in our study had a blood culture quality control study. This data revealed the need to seriously reconsider the applications of blood cultures during the laboratory stage.

In our study, the mean detection time for bacteria considered to be a causative microorganism was 21.4 hours, while for contaminants it was 36.3 hours. Both the literature and the data obtained in our study showed that clinically significant isolates were related to a shorter detection time (15). Therefore, the detection time should serve as an important guiding factor in the determination of contaminants and causative agents.

5.1. Conclusion

Improving blood collection techniques, establishing a phlebotomy team, encouraging venous sampling, and taking more than one blood culture sample can all contribute to reducing the rate of contamination during the pre-analytical phase. It will be appropriate to record time-to-detection values of the blood cultures as well as the number of bottles and detected blood-borne pathogen. During the post-analytical phase, the clinical findings concerning the patients, the number of positive blood culture bottles, and any inflammation markers (i.e., white blood cell count, procalcitonin, and CRP levels) play an important role in determining whether the isolated bacteria is a causative agent or a contaminant.

Footnote

Authors' Contribution: Study concept and design: Mustafa Altindis; acquisition of data: Mustafa Altindis, Mehmet Koroglu, Tuba Dal, Tayfur Demiray; analysis and interpretation of data: Mustafa Altindis, Mehmet Koroglu, and Tayfur Demiray; drafting of the manuscript: Tuba Dal, Mustafa Altindis, and Tayfur Demiray; critical revision of the manuscript: Mustafa Altindis and Oguz Karabay; statistical analysis: Tuba Dal; administrative, technical, and material support: Mustafa Altindis, Tayfur Demiray and Ali Riza Atasoy; study supervision: Mustafa Altindis.

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