

Bacteriological Profile and Antibiotics used for Septic Patients in Karbala, Iraq

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Abstract

Background: Sepsis is an issue of globular health care, characterized by complete inflammation of the body in response to microbial infection, which results in organ malfunction. This becomes a chronic patient complication. Therefore, sepsis may be a diverse and combined disease with varying etiology, severity, and treatment plan. **Objectives:** The purpose of this study was to demonstrate the existence of exposure and the resistance of microorganisms to the range of antibiotics in subjects of the local community. **Results:** Among 11 species of bacteria isolated from a variety of specimens, *Escherichia coli* (22.1%) and *Staphylococcus aureus* (20.3%) were the most common organism found over the subjects had been collected. The most sensitive antibiotics for Gram-negative *E. coli* were colistin 97%, imipenem 89%, meropenem 88%, and amikacin 79%, while the highest resistant rate was benzylpenicillin 93%, and oxacillin 86%. Methicillin-resistant *S. aureus* (MRSA) prevalence was 90%, vancomycin-resistant *S. aureus* (VRSA) 31%. **Conclusion:** Because of the high incidence of VRSA and MRSA, clinicians should become more vigilant in using vancomycin and another important antibiotic. Moreover, maintain this useful drug to be used for a prolonged time with good efficacy.

Keywords: Antibiotics, bacterial profile, methicillin-resistant *Staphylococcus aureus*, sepsis, vancomycin-resistant *Staphylococcus aureus*

INTRODUCTION

Studying the bacterial profile of sepsis patients has a significant importance since sepsis mortality rates ranged from 20% of mild to moderate cases to over 60% in serious patient cases.

[1] An approximate 19 million cases worldwide per year.[2] That meaning sepsis causes one death every 34 s.[3] When the patient receives sufficient antimicrobial therapy within the 1st h of diagnosis, the probability of survival might be approximately 80%; however, it might be decreased by 7.6% every hour afterward. On the other hand, patients who undergone inadequate antimicrobial therapy, could be in risk of survival is five times lower.[4]

Drug resistance is very relevant as it progresses, about 700,000 patients die from drug-resistant infections annually worldwide. Resistant antibiotics include multidrug-resistant tuberculosis and methicillin-resistant *Staphylococcus aureus* (MRSA),[5,6] whereas resistance to carbapenems is increased due to the emergence of carbapenemase bacteria, beta-lactamases that inactivate imipenem and meropenem.[7,8] Recently, colistin resistance

through the plasmid-mediated MCR1 gene has been identified, despite concern for the production of entirely drug-resistant Gram-negative species, as well as *S. aureus* being highly resistant to vancomycin (VRSA), increasing chloroquine resistance globally, and some *Plasmodium falciparum* strains have developed resistance to almost all antimalarial drugs.[9]

Many studies have already shown that ineffective experimental antibiotic regimens have adverse effects including high antibiotic resistance thresholds and elevated morbidity and mortality.[10,11]

Locally, no prior researches were performed to classify the type of bacteria in critical patients, no data on drug sensitivity and resistance. Therefore, clinicians were treated their patients

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based on evidence from other countries such as USA and UK guideline for common bacteria and AB use. Iraq also has biosafety level 2 which mean there is no culture for many micro-organisms like *Brucella*, TB.

MATERIALS AND METHODS

Study design and population

A hospital-based cross-sectional study has been conducted among three main hospitals in Karbala province, Iraq, which includes one public hospital (Al-Hussain Medical City) and two private hospitals (Al-Kafeel and Al-Hujja Hospital) for 2 years between August 2017 and August 2019. Periodically for 2 years in the main three hospitals in Kerbala city, Iraq, all study population were collected. Subjects criteria were included: most patients with sepsis and most patients who had fever for a long time without an apparent diagnosis.

Sampling procedures and sample size

The sampling method was based on the consecutive sampling technique. Cross-sectional studies were performed to examine the antibiotic susceptibility with a wide range of bacteria. Collection of blood cultures was performed as soon as possible after the onset of clinical features of sepsis, before the administration of antibiotics. Since the aim of this study was to study causative micro-organism and the antimicrobial drugs which play important role in improving patient health. The sample size of the study was calculated based on the Thompson equation. The samples required on confidence (95%) and margin of error (5%) were to be 291 samples.

Inclusion and exclusion criteria

Inclusion criteria included all patients complaining from fever for more than 2 weeks without diagnosis, the patient who have deterioration of his/her condition, and patients were suspected to have sepsis according to the NICE or NHS guideline were included in the study. Exclusion criteria included samples excluded from the study were for patients under 16 years of age, non-Iraqi nationality, contaminated samples, and nonblood samples such as wound, urine, cerebrospinal fluid, or sputum.

Ethical consideration

The study was conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki. Ethical clearance was reviewed before the start of the research which was received from the Al Hussein Teaching Medical City Ethical Committee. Samples were obtained on request from the hospital.

Blood sample collection

Blood culture was performed by drawing the blood sample (8–10 ml) from the vein using the aseptic technique. Samples were injected into the bottle tube containing the patient's name a liquid known as broth. The broth was used in order to enhance the bacteria to grow. The inoculated bottles were transported as quickly as possible to the clinical microbiological laboratory for further tests mostly within 1 h. System of continuous-monitoring blood culture was used. Each sample was analyzed by BACT/

ALERT 3D (manufactured by Biomerieux). All blood cultures were loaded in the analyzer for up to 5–8 days, the negatives results remained in the machine for up to 7 days, and the final results were released. As the microorganism grows inside the bottle then release special light. Samples were taken as regular steps to detect what kind of bacteria there. Positive blood cultures were obtained, and biochemical tests were performed using the VITEK compact 2 machine. The necessary biochemical tests were performed. Such as indole methyl red, coagulase, catalase test, Voges–Proskauer, urease, oxidase, and sugar fermentation reaction.^[12,13]

Quality control in blood culture collection

Quality control in blood culture collection involves following procedures exactly to prevent inaccurate results that could delay patient treatment. The rate of false-positive and false-negative blood cultures is monitored by the Lab's Microbiology Department.

Antibacterial susceptibility testing

The examination of antibiotic susceptibility by antibiotic disks was according to the program described by the Committee for Clinical Laboratory Standards.^[14]

Data analysis and interpretation

The descriptive statistical calculations were performed using Microsoft Office Excel 2016 software. Distributions of the antibiotics response were found to be skewed; hence, nonparametric analysis using percentiles was used in measuring the variation.

RESULTS

The total number of the samples was 433 which were collected locally from the main hospitals in Karbala city following the pattern: 249 samples from Imam Hussain medical city, 134 samples from Al-Kafeel hospital, and 50 samples from Al-Hujja hospital. Gender distribution was 298 male samples (68.9%) and 135 female samples (31.1%).

Isolation of Bacteria: Results were demonstrated 11 species of bacteria which were isolated from the total number of specimens as shown in Figure 1.

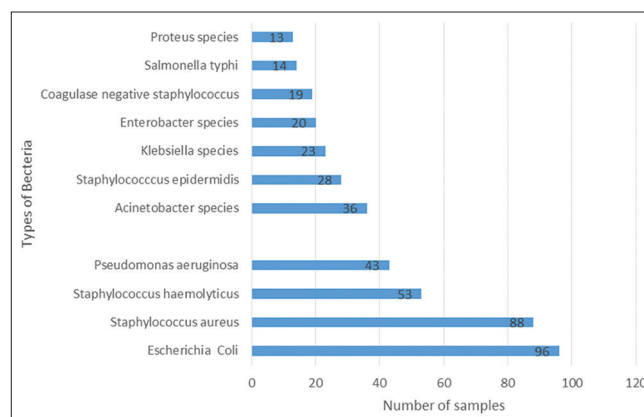


Figure 1: Types of bacteria in blood samples

Antibiotic sensitivity percentages are shown in Table 1 based on common effective treatments for bacteremic or sepsis patients, whereas Table 2 shows the most resistance bacteria susceptibility.

In terms of antibiotic sensitivity, treatment of patients who presented with bacteremia or sepsis, results revealed that the best antibiotic to use was empirical therapy such as teicoplanin which revealed a high sensitivity (92%) to the most prevalent bacteria followed by linezolid 91%, vancomycin 88%, nitrofurantoin 83%, colistin 81%, meropenem 76%, imipenem 59%, amikacin 50% as shown in Table 2. Moreover, benzylpenicillin was the most antibiotic to be avoided due to higher resistance (93%) accompanied by oxacillin 86%, erythromycin 81%, tetracycline 70%, and clindamycin 50%.

Escherichia coli (the most specific Gram-negative organism) antibiotic sensitivity was observed with colistin 97%; imipenem 89%; amikacin 79%; gentamicin 68%; piperacillin/tazobactam 51%; moxifloxacin 50%; levofloxacin 41%; ciprofloxacin 41.1%; septrin 30%; ceftriaxone 25%; and cefepime 6.8%. While *Pseudomonas aeruginosa* showed less sensitivity to most of the antibiotics: colistin 54%, meropenem 40%, ciprofloxacin 40%, piperacillin 33%, imipenem 27%, amikacin 27%, and it was 100% resistant to gentamicin, levofloxacin, ampicillin/sulbactam, and ceftriaxone.

DISCUSSION

Among 1229 blood samples of patients suspected of having septicemia over the study period, 796 samples (64.8%) were negative in culture and 433 samples (35.2%) were positive in bacteria culture. Positive test outcomes were higher in number than in other studies including Hassan *et al.* (8.3%) in Baghdad, Gohel *et al.* (9.2%), Mehta *et al.* (9.9%), and Kotgire and Hatkar (19.2%), respectively.^[15-18] That might be through delay in sending blood culture samples due to the high price of this test for most patients of low socioeconomic status or it might arise by diagnostic time due to the doctor himself.

The study included 433 samples as a total number of patients: 298 males (68.9%) and 135 females (31.1%), which was very similar to Gohel *et al.* Their study included 67.5% males and 32.5% females, but it was different from the study of Hassan *et al.* which included 53.8% males and 46.2% females, Hussein *et al.* 66.5% male and 33.5% female and Salari (87%) male and (13%) female^[15,16,19-21] respectively.

The total species were 27 types of bacteria, the most common types was 11 species accounting for 99% of the bacteria shown in Figure 1 from the Gram-positive species bacteria accounting for 47.8%, from these Gram-positive species, *S. aureus* (20.3%) was the predominant while the Gram-negative bacteria accounting for 51.8%. *E. coli* (33.1%) was the predominated as compared to other studies. The incidence of Gram-positive in the current study was lower than that obtained by Kotgire and Hatkar (53.9%) who showed that the frequency of Gram-positive and Gram-negative bacteria was 53.9% and

Table 1: Antibacterial susceptibility to bacteria according to the most sensitive type

Susceptibility to bacteria	Antibiotics										
	Teicoplanin	linezolid	Vancomycin	Nitrofurantoin	Colistin	Meropenem	Imipenem	Moxifloxacin	Gentamicin	Levofloxacin	Fosfomycin
Sensitive (%)	92	91	88	83	81	76	70	59	53	50	49
Resistant (%)	8	9	12	14	18	23	26	39	45	47	51
Intermediate (%)	0	0	0	3	1	1	4	2	2	3	0
Total number of sample	112	160	148	121	80	109	108	127	224	176	104

Table 2: Antibacterial susceptibility to bacteria according to most resistance types

Susceptibility to bacteria	Antibiotics								
	Benzylpenicillin	Oxacillin	Erythromycin	Tetracycline	Clindamycin	Fosfomycin	Cefepime	Ceftazidime	Septrin
Sensitive (%)	93	86	81	70	59	51	49	48	48
Resistant (%)	6	14	18	30	39	49	17	32	46
Intermediate (%)	1	0	1	0	2	0	34	20	6
Total number of sample	120	77	149	139	155	104	102	56	224

46.1%, respectively.^[18] Comparing to other studies,^[15,22,23] their findings revealed that *S. aureus* was the most frequent causative bacteria in their samples.

Results also showed that the prevalence of Gram-negative (*E. coli*, *Klebsiella*, and *Proteus*) was 30%, showing an agreement with other research including Hassan *et al.* (34.4%) and Ahmed and Hussein (37%).^[15,24] In comparison, the prevalence of *Pseudomonas aeruginosa* was 9.9%, less than Hassan *et al.* 12.3% and more than Kumar *et al.* 7.6% and Gupta *et al.* 5.9%,^[15,25,26] while *Salmonella typhi* was 3.2% which is less than Kotgire and Hatkar (13.88%).^[18]

In comparison with similar studies performed, locally, the most microorganism in burning patients was *P. aeruginosa*, which showed 58% resistant to imipenem, 66% to meropenem, 86% to piperacillin, 51% to ceftazidime, 43% to aztreonam, 47% to ciprofloxacin, and gentamicin 91%,^[27] whereas in our study, the *P. aeruginosa* sensitivity toward colistin was 54%, meropenem 40%, ciprofloxacin 40%, piperacillin 33%, imipenem 27%, amikacin 27%, and there was 100% resistant to gentamicin, levofloxacin, ampicillin/sulbactam, and ceftriaxone. This indicates that meropenem remains 40% sensitive in the same type of activity and 66% resistant in that 7-year cycle (2012–2019) While imipenem sensitivity decreased from 63% in Shilba *et al.*,^[27] 42% in Abboud *et al.*^[28] to only 27% in our study, and that due to the uses increase. Piperacillin becomes more sensitive from 14% in 2012^[27] to 33% in 2019, while gentamicin stays on the same level of activity. It was 91% resistant in 2012,^[27] while it showed 100% in our study.

On the other hand, regarding *E. coli*, their susceptibility to imipenem and amikacin has increased from 77% to 89% and from 66% to 79%, respectively, compared with Abboud *et al.*^[28] and this study.

Furthermore, results were shown that MRSA prevalence of all forms of *Staphylococcus* was 90%, very high compared to Gohel *et al.* 70%,^[16] Malaysia was 17% in 1986 while becomes 44% in 2007. Europe varies from 0.9% in the Netherlands to 56% in Romania, while the prevalence of 15 studies indicated (13%–74% worldwide). The reasons behind this may be attributable to overuse of antistaph or MRSA treatment, which is performed depending on cefoxitin resistance.

The sensitivity of *S. aureus* to antibiotics was as following: linezolid 90.7%, teicoplanin 71.1%, vancomycin 68.4% while clindamycin was 33.33% of Gram-positive organism. Moreover, VRSA (vancomycin-resistant *S. aureus*) was 31.6%

higher than in Gohel *et al.* 21.6%.^[16] Results also shown a high positive blood culture was for *Staphylococcus epidermidis* and *Staphylococcus haemolyticus*.

CONCLUSION

Growing the incidence of MRSA in our society has led to more concern and a strict antimicrobial policy being awarded for using without medical guidance. Relatively, increased MRSA and VRSA incidence compared to the other research could be outlined locally based on the following reasons: first, improper diagnosis: most physicians involved in treating patients with fever for more than 2 weeks without any other indications were treated as having sepsis without following the sepsis guideline. That leads strongly to incorrect treatment line and raises the incidence of the study case. Second, patients and physicians are entirely responsible for the incomplete course of treatment and for the inappropriate dosage of antibiotics, which will also increase the case of the study. Third, the most crucial cause of high incidence vancomycin resistance is the availability in health ministry stores and the absence of other antibiotics for MRSA. Finally, it would be extremely important to the Ministry of Health to provide other antibiotics that act against MRSA, such as teicoplanin, linezolid, or colistin. Moreover, to make strict rules for internists to followed the patient's diagnostic and treatment guidelines.

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Conflicts of interest

There are no conflicts of interest.

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