

The Most Predominant Gram Negative Bacteria in a Public Hospital

ABSTRACT

Aim: the aim of this study was to explore the occurrence of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in a hospital in Al-Kharj.

Methodology: The study included the review of antibiogram results to find the number and percentage of these bacteria in 4 years, 2015-2018.

Results: The total number of bacterial cultures in the 4 years was 3327. Most of these cultures were for gram negative bacteria (81.06%). Regarding gram negative bacteria in the 4 years, the most common was *Escherichia coli* (represents 19.30% of the total bacteria and 23.80% of gram negative bacteria).

Conclusion: It can be concluded that the most prevalent gram negative bacteria were *Escherichia coli*, *Pseudomonas aeruginosa* and *Klebsiella pneumonia*. It is important to conduct more researches to know the prevalence of these bacteria, to know their resistance rate to help health care providers to prescribe and dispense antibiotics wisely.

Keywords: *Acinetobacter baumannii*, *Escherichia coli*, *Gram negative bacteria*, *Klebsiella pneumonia*, *prevalence*, *Pseudomonas aeruginosa*.

INTRODUCTION

A worldwide increase in the number of infections caused by gram-negative bacteria has occurred in current years, with infections caused by gram-negative organisms often more prevalent than gram-positive infections in numerous settings [1]. Gram-negative bacteria cause infections including bloodstream infections, pneumonia, meningitis, and wound or surgical site infections in healthcare settings. Gram-negative infections include those caused by *Pseudomonas aeruginosa*, *Acinetobacter*, *Escherichia coli* and *Klebsiella* as well as many other less common bacteria [2].

In the United States, it is estimated that as many as 2 million patients yearly develop a bacterial infection (caused by gram positive and/or gram-negative bacteria) that is resistant to antibiotics and that more than 23,000 deaths are associated with these infections [3]. A majority of these deaths are related to gram-negative infections, particularly health care–acquired infections caused by extended-spectrum b-lactamase-producing *Enterobacteriaceae*, carbapenem-resistant *Enterobacteriaceae*, multidrug resistance *Pseudomonas aeruginosa* and multidrug resistance *Acinetobacter baumannii* [3]. Centers for Disease Control and Prevention reported that these multidrug resistance gram negative bacteria such as carbapenem-resistant *Enterobacteriaceae* and *Acinetobacter* spp in particular are increasing in prevalence [4].

Escherichia coli normally resides in the intestinal flora of humans and other warm-blooded animals. It is ubiquitous in the environment and has been used as an indicator of fecal contamination to assess the quality and safety of water [5]. Although most *Escherichia coli* strains are harmless, certain strains are pathogenic and cause several diseases such as bloody diarrhea, watery diarrhea, meningitis, urinary tract infection and sepsis which can lead to death [6,7]. *Pseudomonas* is a type of bacteria that is found commonly in the environment, like in water and in soil. There are many different types of *Pseudomonas*, *Pseudomonas aeruginosa* is the organism that most often causes infections in humans. It can cause infections in the lungs, blood or other parts of the body after surgery [8].

Klebsiella species rank among the top 10 bacteria causing hospital-acquired infections, and it is one of the most common bacteria isolated in the intensive care unit (ICU). *Klebsiella pneumoniae* is a gram-negative facultative anaerobic bacillus that is a member of the *Enterobacteriaceae* family. It is found in the respiratory tract

and stools of about 5% of normal individuals. It is responsible for about 1% of bacterial pneumonia [9-11].

Klebsiella pneumoniae is responsible for a significant proportion of soft tissue infections, pneumonia and urinary tract infections [10]. *Acinetobacter baumannii* can cause infections in the blood, lungs, urinary tract, or in wounds in other parts of the body. It can also “colonize” or live in a patient without causing infections or without symptoms, especially in respiratory secretions or open wounds [12]. In the United States, *Acinetobacter* infections rarely occur outside of healthcare settings. However, people who have weakened immune systems or who have chronic diseases may be more susceptible [12].

These 4 bacteria are usually the most prevalent gram negative bacteria. This study aimed to explore the prevalence of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria.

METHODOLOGY

This retrospective study was conducted in a public hospital in Al-Kharj to explore the occurrence of the most prevalent gram negative bacteria. The study included the review of antibiogram results to find the number and percentage of these bacteria in 4 years, 2015-2018.

The data were collected from microbiology laboratory in the hospital after receiving the acceptance from the hospital management. The data were represented as frequency and percentage of gram positive and gram negative bacteria in the 4 years and frequency and percentage of the most prevalent gram negative bacteria.

This study was approved by institutional review board (IRB) committee of Saudi Ministry of Health and after receiving the acceptance from the public hospital. The study was approved with IRB log number of 18-474E.

RESULTS AND DISCUSSION

The number of bacterial cultures in 2015 was 897. Most of these cultures were for gram negative bacteria (81.83%).

Table 1 shows the number of gram negative and gram positive bacteria in 2015.

Table 1. Number of gram negative and gram positive bacteria in 2015

Bacteria	Number	Percentage
Gram negative bacteria	734	81.83%
Gram positive bacteria	163	18.17%
Total	897	100.00%

Regarding gram negative bacteria in 2015, the most common was *Escherichia coli* (represents 21.63% of the total bacteria and 26.43% of gram negative bacteria) followed by *Pseudomonas aeruginosa* (represents 18.62% of the total bacteria and 22.75% of gram negative bacteria). Table 2 showed the number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2015

Table 2. Number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2015

Bacteria	Number	Percentage*
<i>Escherichia coli</i>	194	21.63%
<i>Klebsiella pneumoniae</i>	94	10.48%
<i>Pseudomonas aeruginosa</i>	167	18.62%
<i>Acinetobacter baumannii</i>	78	8.70%

*The percentage of bacteria is calculated as number of bacteria divided by the total bacteria (gram positive and gram negative bacteria) so the sum of the percentage was less than 100%.

The number of bacterial cultures in 2016 was 920. Most of these cultures were for gram negative bacteria (82.61%).

Table 3 shows the number of gram negative and gram positive bacteria in 2016.

Table 3. Number of gram negative and gram positive bacteria in 2016

Bacteria	Number	Percentage
Gram negative bacteria	760	82.61%
Gram positive bacteria	160	17.39%
Total	920	100.00%

Table 4 showed the number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2016. Regarding gram negative bacteria in 2015, the most common was *Pseudomonas aeruginosa* (represents 19.57% of the total bacteria and 23.68% of gram negative bacteria) followed by *Escherichia coli* (represents 16.09% of the total bacteria and 19.47% of gram negative bacteria).

Table 4. Number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2016.

Bacteria	Number	Percentage*
<i>Escherichia coli</i>	148	16.09%
<i>Klebsiella pneumoniae</i>	119	12.93%
<i>Pseudomonas aeruginosa</i>	180	19.57%
<i>Acinetobacter baumannii</i>	100	10.87%

*The percentage of bacteria is calculated as number of bacteria divided by the total bacteria (gram positive and gram negative bacteria) so the sum of the percentage was less than 100%.

The number of bacterial cultures in 2017 was 765. Most of these cultures were for gram negative bacteria (83.79%).

Table 5 shows the number of gram negative and gram positive bacteria in 2017.

Table 5. Number of gram negative and gram positive bacteria in 2017

Bacteria	Number	Percentage
Gram negative bacteria	641	83.79%
Gram positive bacteria	124	16.21%
Total	765	100.00%

Table 6 showed the number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2017. Regarding gram negative bacteria in 2017, the most common was *Escherichia coli* (represents 18.17% of the total bacteria and 21.68% of gram negative bacteria) followed by *Pseudomonas aeruginosa* (represents 17.39% of the total bacteria and 20.75% of gram negative bacteria).

Table 6. Number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2017

Bacteria	Number	Percentage*
<i>Escherichia coli</i>	139	18.17%
<i>Klebsiella pneumonia</i>	112	14.64%
<i>Pseudomonas aeruginosa</i>	133	17.39%
<i>Acinetobacter baumannii</i>	98	12.81%

*The percentage of bacteria is calculated as number of bacteria divided by the total bacteria (gram positive and gram negative bacteria) so the sum of the percentage was less than 100%.

The number of bacterial cultures in 2018 was 745. Most of these cultures were for gram negative bacteria (75.44%).

Table 7 shows the number of gram negative and gram positive bacteria in 2018.

Table 7. Number of gram negative and gram positive bacteria in 2018

Bacteria	Number	Percentage
Gram negative bacteria	562	75.44%
Gram positive bacteria	183	24.56%
Total	745	100.00%

Table 8 showed the number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2018. Regarding gram negative bacteria in 2018, the most common was *Escherichia coli* (represents 21.61% of the total bacteria and 28.65% of gram negative bacteria) followed by *Klebsiella pneumoniae* (represents 17.99% of the total bacteria and 23.84% of gram negative bacteria).

Table 8. Number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in 2018

Bacteria	Number	Percentage*
<i>Escherichia coli</i>	161	21.61%
<i>Klebsiella pneumonia</i>	134	17.99%
<i>Pseudomonas aeruginosa</i>	105	14.09%

<i>Acinetobacter baumannii</i>	80	10.74%
--------------------------------	----	--------

*The percentage of bacteria is calculated as number of bacteria divided by the total bacteria (gram positive and gram negative bacteria) so the sum of the percentage was less than 100%.

The total number of bacterial cultures in the 4 years was 3327. Most of these cultures were for gram negative bacteria (81.06%). Table 9 shows the number of gram negative and gram positive bacteria in the 4 years.

Table 9. Total number of gram negative and gram positive bacteria in the 4 years (2015-2018)

Bacteria	Number	Percentage
Gram negative bacteria	2697	81.06%
Gram positive bacteria	630	18.94%
Total	3327	100.00%

Regarding gram negative bacteria in the 4 years, the most common was *Escherichia coli* (represents 19.30% of the total bacteria and 23.80% of gram negative bacteria) followed by *Pseudomonas aeruginosa* (represents 17.58% of the total bacteria and 21.69% of gram negative bacteria). Table 10 showed the number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in in the 4 years.

Table 10. Total number of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* bacteria in the 4 years (2015-2018)

Bacteria	Number	Percentage*
<i>Escherichia coli</i>	642	19.30%
<i>Klebsiella pneumoniae</i>	459	13.80%
<i>Pseudomonas aeruginosa</i>	585	17.58%
<i>Acinetobacter baumannii</i>	356	10.70%

*The percentage of bacteria is calculated as number of bacteria divided by the total bacteria (gram positive and gram negative bacteria) so the sum of the percentage was less than 100%.

The present study showed that gram negative bacteria were more common than gram positive bacteria and that the most prevalent gram negative bacteria were *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumonia* and *Acinetobacter baumannii*. Rotimi et al conducted a study about the prevalence of gram negative bacteria but in intensive care units and stated that the most common bacterial isolates in Jeddah versus Kuwait intensive care units were *Pseudomonas aeruginosa* (26%, 26%), *Escherichia coli* (23%, 3%), *Klebsiella pneumoniae* (20%, 17%), inducible *Enterobacteraeae* group (17%, 14%), and *Acinetobacter* spp. (9%, 33%) [13]. A systematic review and meta-analysis was conducted by Aliyu et al and stated that among nursing home residents, *Escherichia coli* accounted for the largest proportion of multidrug-resistant gram-negative bacterial isolates [14]. Moreover, Zare et al reported that among gram-negative bacteria isolated from patients with urinary tract infections, the most

commonly isolated bacteria in the urine included *Escherichia coli* (45%), *Enterobacter* (12%), *Klebsiella* (3.14%), and *Pseudomonas* isolates (1.22%) [15]. Additionally, saber et al and Gajdács et al reported that *Escherichia coli* has been documented to be the most common pathogen associated with urinary tract infections in many countries causing both community- and hospital-acquired UTI [16,17].

Al-Saadi et al reported that the most frequent identified bacterial species in open fractures were *Staphylococcus aureus* (23.52%) followed by *Acinetobacter* spp (19.32%) ,then *Escherichia coli* (14.28%), *Pseudomonas* spp (11.76%), *Enterobacter* spp (9.24%), *Klebsiella* spp (6.72%) [18]. Deka et al reported that regarding gram-negative bacteria causing surgical site infection in a tertiary care hospital, *Escherichia coli* (22.4%) and *Klebsiella* species (20.6%) were the predominant isolated bacteria followed by *Staphylococcus* species (18.4%), *Pseudomonas* species (12.3%), and *Enterococcus* species (6.6%) [19]. Wen et al stated that data from the National Nosocomial Infection Surveillance System (NNISS) in mainland China indicated that *Pseudomonas aeruginosa* ranked top among pathogens identified from the lower respiratory tract, at 12.82% from 1999 to 2001, 12.31% during the period from 2002 to 2004, and 13.37% from 2005 to 2007 [20].

LIMITATIONS

The main limitation in the study was that the diagnosis is not available so the types of infections were not identified and also the setting is not determined, for example if these cultures isolated from ICU or inpatients.

CONCLUSION

The present study showed that gram negative bacteria were more common than gram positive bacteria and that the most prevalent gram negative bacteria were *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Acinetobacter baumannii*. It is important to know the prevalence of infections caused by these bacteria and also to know their resistance rate. More researches are needed to prepare stratified antibiogram that will help health care providers to prescribe and dispense antibiotics wisely.

References

1. Vincent JL, Rello J, Marshall J, Silva E, Anzueto A, Martin CD, et al. International Study of the Prevalence and Outcomes of Infection in Intensive Care Units. *JAMA*. 2009;302(21): 2323-9.

2. CDC. Gram negative bacteria. Available at: <https://www.cdc.gov/hai/organisms/gram-negative-bacteria.html>.
3. CDC. Antibiotic resistance threats in the United States. Available at: <http://www.cdc.gov/drugresistance/threat-report-2013/pdf/arthreats-2013-508.pdf>.
4. CDC. Multi-site gram-negative bacilli surveillance initiative. Available at: <http://www.cdc.gov/hai/eip/mugsi.html>.
5. EPA. Ambient water quality criteria for bacteria. Washington, D.C.: U.S. EPA, Office of Water Regulations and Standards Division; 1986.
6. Nataro JP, Kaper JB. Diarrheagenic Escherichia coli. *Clin Microbiol Rev.* 1998;11(1):142–201.
7. Gyles CL. Shiga toxin-producing Escherichia coli: an overview. *J Anim Sci.* 2007;85(13 Suppl):E45–E62.
8. CDC. Pseudomonas. Available at: <https://www.cdc.gov/hai/organisms/pseudomonas.html>.
9. Spagnolo AM, Orlando P, Panatto D, Perdelli F, Cristina ML. An overview of carbapenem-resistant Klebsiella pneumoniae: epidemiology and control measures. *Rev Med Microbiol.* 2014;25:7-14.
10. Morse AM, Brooks GF, Carroll KC, Butel JS, Melnick T. Enteric gram-negative rods (Enterobacteriaceae). Melnick & Adelberg's Medical Microbiology, 26th Edition. McGraw-Hill Education, New York. 2013;229-244.
11. Sanchez GV, Master RN, Clark RB, Fyyaz M, Duvvuri P, Ekta G, et al. Klebsiella pneumoniae Antimicrobial Drug Resistance, United States, 1998–2010. *Emerg Infect Dis.* 2013;19(1):133-136.
12. CDC. Acinetobacter. Available at: <https://www.cdc.gov/hai/organisms/acinetobacter.html>.
13. Rotimi VO, Al-Sweih NA, Feteih J. The prevalence and antibiotic susceptibility pattern of gram-negative bacterial isolates in two ICUs in Saudi Arabia and Kuwait. *Diagn Microbiol Infect Dis.* 1998;30(1):53-59.
14. Aliyu S, Smaldone A, Larson E. Prevalence of multidrug-resistant gram-negative bacteria among nursing home residents: A systematic review and meta-analysis. *Am J Infect Control.* 2017;45(5):512-518.
15. Zare F, Mohammadzadeh Rostami F, Shahsafi M. Prevalence and pattern of antibiotic resistance of gram-negative bacteria isolated from urinary tract infections in patients referring to Neka Laboratories-Iran. *Int J BioMed Public Health.* 2018;1(1):30-36.
16. Sabir S, Anjum AA, Ijaz T, Ali MA, Rehman MU, Nawaz M. Isolation and antibiotic susceptibility of E. coli from urinary tract infections in a tertiary care hospital. *Pak J Med Sci.* 2014;30(2):389–392.
17. Gajdács M, Ábrók M, Lázár A, Burián K. Comparative Epidemiology and Resistance Trends of Common Urinary Pathogens in a Tertiary-Care Hospital: A 10-Year Surveillance Study. *Medicina.* 2019;55(7):356.
18. Al-Saadi AGM, Abbas AF, Fazaa SA. The Prevalence of the Gram Positive and Gram Negative Bacteria in Open Fractures and their Resistance Profiles to Antimicrobial Agents. *JGPT.* 2017;12(9):69-76.

19. Deka S, Kalita D, Mahanta P, Baruah D. High Prevalence of Antibiotic-Resistant GramNegative Bacteria Causing Surgical Site Infection in a Tertiary Care Hospital of Northeast India. *Cureus*. 2020;12(12):e12208.
20. Wen XM, Ren N, Wu AH, Xu XH. Distribution of pathogens causing nosocomial infection monitored by national nosocomial infection surveillance system and changing trend. *CJN*. 2011;21(2011):350-355.

UNDER PEER REVIEW