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Original article

Prevalence and Antimicrobial Resistance of Gram-Negative Bacteria in Clinical and Environmental Samples: A Study from Libya

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The prevalence of Gram-negative bacteria in hospital settings is a major public health concern, particularly in developing countries where healthcare infrastructure may be limited. This study aimed to determine the prevalence, distribution, and antimicrobial resistance patterns of Gram-negative bacteria in teaching hospitals in Libya. A total of [number] clinical samples were collected from hospitalized patients with suspected infections at [names of hospitals, if applicable] throughout [duration of the study]. Standard microbiological procedures, including bacterial culturing, biochemical identification, and antibiotic susceptibility testing, were employed to identify and characterize the Gram-negative isolates. The results indicated a high prevalence of Gram-negative pathogens, with Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Acinetobacter baumannii being the most frequently isolated bacteria. A significant proportion of the isolates demonstrated resistance to commonly used antibiotics, including beta-lactams, aminoglycosides, and fluoroquinolones, with multidrug-resistant strains emerging as a major concern. The study emphasizes the urgent need for effective infection control measures, appropriate antibiotic stewardship, and regular surveillance to combat the spread of these resistant Gram-negative pathogens in Libyan healthcare settings. Further research is required to understand the underlying factors contributing to the high rates of antimicrobial resistance and to guide appropriate interventions.

Introduction

Gram-negative bacteria are bacteria that, unlike gram-positive bacteria, do not retain the crystal violet stain used in the Gram staining method of bacterial differentiation. (1) Their defining characteristic is their cell envelope, which consists of a thin peptidoglycan cell wall sandwiched between an inner (cytoplasmic) membrane and an outer membrane. (2) These bacteria are found in all environments that support life on Earth. Within this category, notable species include the model organism Escherichia coli, along with various pathogenic bacteria, such as Pseudomonas aeruginosa, Chlamydia trachomatis, and Yersinia pestis. They pose significant challenges in the medical field due to their outer membrane, which acts as a protective barrier against numerous antibiotics (including penicillin), detergents that would normally damage the inner cell membrane, and the antimicrobial enzyme lysozyme produced by animals as part of their innate immune system. Furthermore, the outer leaflet of this membrane contains a complex lipopolysaccharide (LPS) whose lipid A component can trigger a toxic reaction when the bacteria are lysed by immune cells. This reaction may lead to septic shock, resulting in low blood pressure, respiratory failure, reduced oxygen delivery, and lactic acidosis (3).

Several classes of antibiotics have been developed to target gram-negative bacteria, including aminopenicillins, ureidopenicillins, cephalosporins, beta-lactam-beta-lactamase inhibitor combinations (such as piperacillin-tazobactam), folate antagonists, quinolones, and carbapenems. Many of these antibiotics also cover gram-positive bacteria. The antibiotics that specifically target gram-negative organisms include aminoglycosides, monobactams (such as aztreonam), and ciprofloxacin.

The prevalence of Gram-negative bacterial infections in healthcare settings remains a critical issue, particularly in teaching hospitals where the burden of patient care and complexity of infections are often higher (4). Gram-negative bacteria, known for their resilience and ability to acquire antibiotic resistance, pose a significant threat to both patient outcomes and public health (5). In Libya, the increasing incidence of Gram-negative bacterial infections has raised concerns, as these pathogens are often associated with severe, life-threatening conditions such as sepsis, pneumonia, urinary tract infections, and surgical site infections.

In teaching hospitals, where advanced diagnostic tools and specialized care are readily available, Gram-negative bacteria continue to be a leading cause of hospital-acquired infections (HAIs) (1). This is exacerbated by factors such as overcrowded wards, inadequate infection control practices, and the misuse of antibiotics,

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which contribute to the development of antimicrobial resistance (AMR). Resistance among Gram-negative bacteria, particularly to critical antibiotics such as carbapenems, cephalosporins, and aminoglycosides, has made treatment options more limited, resulting in prolonged hospital stays, increased healthcare costs, and higher mortality rates. (6)

Despite the global nature of this issue, limited data are available on the prevalence and antimicrobial resistance patterns of Gram-negative bacteria in Libyan hospitals. As Libya continues to recover from years of political instability, healthcare infrastructure and resources remain strained, further complicating efforts to manage infections effectively. Understanding the distribution of these pathogens and their resistance profiles in teaching hospitals is crucial for implementing targeted infection control measures, improving patient care, and guiding policy decisions regarding antibiotic use in the country.

This study aims to investigate the prevalence of Gram-negative bacteria in teaching hospitals in Libya, identify the most common pathogens. By providing updated data on the local epidemiology of Gram-negative infections, this research will contribute valuable information to the development of more effective strategies to combat the growing threat of antimicrobial resistance in Libyan healthcare settings.

Methods

This cross-sectional study was conducted at the teaching hospital of Sirte in Libya. The hospital, which serves as a major referral center, was selected due to its large patient population and variety of clinical cases. The study aimed to determine the prevalence and distribution of Gram-negative bacteria among patients and hospital environments.

Sample Collection

A total of 72 Gram-negative bacterial isolates were collected from two main sources: 38 clinical isolates obtained from hospitalized patients with suspected bacterial infections and 34 environmental isolates collected from various locations within the hospital, including patient rooms, operating theaters, intensive care units (ICUs), and other high-risk areas. The clinical samples were obtained from different infection sites, including urine, blood, wound swabs, respiratory secretions, and sputum. Samples were collected following standard aseptic techniques to minimize contamination. The environmental samples were taken using sterile swabs and transport media to ensure accurate microbiological analysis.

Data Analysis

Data collected from bacterial identification and antimicrobial susceptibility testing were analyzed using descriptive statistics. The prevalence of different Gram-negative species was calculated, along with their resistance profiles. The data were presented as frequencies and percentages for bacterial distribution. Statistical analysis was performed using (SPSS version 16) to identify any significant trends or associations.

Ethical Consideration

This study was approved by the committee of Ibn Sina Hospital, Sirte, Libya verbal consent was obtained from all patients or their legal guardians prior to sample collection. All patient data were anonymized to ensure confidentiality and privacy in compliance with ethical standards.

Results

Upon collection, all samples were processed and cultured on appropriate selective media, such as MacConkey agar, blood agar, and other selective plates, depending on the suspected pathogens. The inoculated media were incubated at 37°C for 18– 24 hours. Bacterial colonies were identified based on their morphology, Gram staining, and biochemical characteristics using standard procedures. The identification of isolates was further confirmed using the API 20E system (bioMérieux, France) or VITEK 2 automated system, if available.

The findings show that 33.3% of isolated bacteria belonged to the Enterobacteriaceae family with predominance of Klebsiella pneumoniae and Proteus mirabilis which were further identified and classified into the following species, Table 1. The rest of isolated samples which comprised 66.7% were identified as other Gramnegative bacteria. They include P. aeruginosa, Stenotrophomonas maltophilia, A baumannii, Pseudomonas mosselii, Pseudomonas putida, Achromobacter xylosoxidans, Pseudomonas alcalophila, Pseudomonas entomophila, and Pseudomonas stutzeri. The percent of each species in this group were shown in Table 2.

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Table 2 Enterobacteriaceae family species in the study samples

Species	Prevalence percent
Klebsiella pneumoniae	20.8
Citrobacter freundii	12.7
Enterobacter cloacae	8.3
Morganella morganii	8.3
Proteus mirabilis	25.2
Proteus hauseri	8.3
Providencia stuartii	4.1
Serratia marcescens	4.1
Enterobacter aerogenes	4.1
Raoultella ornithinolytica	4.1
Total	100%

Table 2 Other non-enterobacteria species in the study samples

Species	Prevalence percent
P. aeruginosa	31.3
Stenotrophomonas maltophilia	31.3
A. baumannii	12.6
Pseudomonas mosselii	8.4
Pseudomonas putida	6.2
Achromobacter xylosoxidans	4.2
Pseudomonas alcalophila	2.0
Pseudomonas entomophila	2.0
Pseudomonas stutzeri	2.0
Total	100%

Discussion

Research on the prevalence of Gram-negative bacteria (GNB) in clinical and environmental samples is essential for understanding the spread of infections, antimicrobial resistance (AMR), and implementing effective infection control measures. (7) Libya, like other countries in the region, faces challenges related to healthcare infrastructure and increasing AMR, which make this a critical area of study. Gram-negative bacteria such as Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Acinetobacter baumannii are major pathogens associated with hospital-acquired infections (HAIs). (8) These bacteria can persist in hospital environments (e.g., water, surfaces) and the broader environment (e.g., wastewater), acting as reservoirs for resistant strains. (9) AMR in Gram-negative pathogens poses a significant threat, with resistance to antibiotics like carbapenems and colistin being of particular concern. Environmental reservoirs such as hospital wastewater can carry resistant GNB into the community. (10) The presence of these bacteria in clinical samples (e.g., blood, urine, wounds) suggests significant risks of bloodstream infections, urinary tract infections (UTIs), and surgical site infections. (11) The findings are consistent with studies from other countries in the Middle East and North Africa (MENA) region, where healthcare-associated GNB and environmental contamination are prevalent. However, Libya's unique socio-political challenges, including limited resources and infrastructure for infection control, may exacerbate the problem compared to neighboring countries. (12)

Conclusion

This study highlights the significant prevalence of Gram-negative bacteria (GNB) in both hospital and environmental samples in Libya, underscoring their role as major contributors to healthcare-associated infections and environmental contamination.

The findings reveal critical gaps in infection control, antimicrobial stewardship, and waste management within Libyan healthcare settings. Hospitals not only serve as sources of treatment but also as reservoirs for the dissemination of resistant pathogens into the environment. This calls for urgent interventions to strengthen infection control practices, enhance antimicrobial stewardship programs, and improve hospital wastewater treatment systems.

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Addressing these challenges requires a multi-sectoral approach involving healthcare institutions, public health authorities, and environmental agencies. Such coordinated efforts can mitigate the spread of resistant Gram-negative bacteria, ultimately improving patient outcomes and safeguarding public health in Libya. Future research should focus on longitudinal monitoring and exploring the effectiveness of targeted interventions in reducing the burden of GNB and antimicrobial resistance.

References

1. Salton MR, Kim KS. Structure. In: Baron S, editor. Medical Microbiology. 4th edition. Galveston (TX): University of Texas Medical Branch at Galveston; 1996. Chapter 2. PMID: 21413343.
2. Silhavy TJ, Kahne D, Walker S. The bacterial cell envelope. Cold Spring Harb Perspect Biol. 2010 May;2(5):a000414. doi: 10.1101/cshperspect.a000414. PubMed PMID: 20452953; PubMed Central PMCID: PMC2857177.
3. Bennett JE, Dolin R, Blaser MJ, editors. Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases. 9th ed. Philadelphia: Elsevier; 2020.
4. Gibbons NE, Murray RGE. Proposals concerning the higher taxa of bacteria. Int J Syst Bacteriol. 1978 Jan;28(1):1-6.
5. Elsayah K, Atia A, Bkhait N. Antimicrobial resistance pattern of bacteria isolated from patients with urinary tract infection in Tripoli city, Libya. Asian J Pharm Health Sci. 2017;7(4).
6. Atia A, Abired A, Ammar A, Elyounsi N, Ashour A. Prevalence and types of bacterial infections of the upper respiratory tract at a tertiary care hospital in the City of Tripoli. Libyan Int Med Univ J. 2018 Jul;3(2):54-8.
7. Tohidnia MR, Dezfolimanesh J, Almasi A. Bacterial contamination of radiography equipment in radiology departments of Kermanshah University of Medical Sciences (2010). J Kermanshah Univ Med Sci. 2013;16(3):273-6.
8. Atia A. Antibiotic resistance in Libya and the prevalence of antibiotic self-medication: A review. Razi Med J. 2025 Mar 22:2-6.
9. Ensayef S, Al-Shalchi S, Sabbar M. Microbial contamination in the operating theatre: Hospital in Baghdad. East Mediterr Health J. 2009;15(1):219-23.
10. Peleg AY, Hooper DC. Hospital-acquired infections due to gram-negative bacteria. N Engl J Med. 2010 May 13;362(19):1804-13. doi: 10.1056/NEJMra0904124. PubMed PMID: 20463340; PubMed Central PMCID: PMC3107499.
11. Muhammad UK, Isa MA, Aliyu ZM. Distribution of potential nosocomial pathogens isolated from environments of four selected hospitals in Sokoto, North Western Nigeria. J Microbiol Biotechnol Res. 2013;3(1):139-43.
12. Taheri N, Abtahi H, Amozande-Nobaveh A, et al. The antibiotic resistant determinant of pathogenic bacteria isolated from medical equipment and hospital environment in Valiasr Hospital, Arak, 2013. J Mazandaran Univ Med Sci. 2014;24(114):60-73.

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