Libyan Open University Journal of Medical Sciences and Sustainability
(https://oujournals.ly/index.php/LOUJMSS/index)
Volume 2 - Issue 1 - 2026 - Pages 38-48

Microbiological and Physicochemical Assessment of Antibiotic-Resistant Bacteria in Hospital Wastewater: A Case Study from Sabratha Teaching Hospital, Libya

[1] Libyan Center for Water and Soil Research, Studies and Desertification Control – Sabratha
[2-5]Department of Environmental Engineering ,Faculty of Engineering ,Sabratha University , Libya.
[6] Department of Biology, Faculty of Education, Bani Waleed University, Libya
[*]**Email for reference researcher:** adelbanana@yahoo.com

التقييم الميكروبيولوجي والفيزيائي والكيميائي للبكتيريا المقاومة للمضادات الحيوية في مياه الصرف الصحي للمستشفيات: دراسة حالة من مستشفى صبراتة التعليمي، ليبيا

هناء سليمان زهمول [1]، عادل عاشور بنانه [2] [*]، علي ضوء عبعوب[3] ، جمعة السيد الطاهر محمد [4]، ريمة محمد سلام[5] ، آمنة علي الحداد[6]
[1] المركز الليبي لابحاث ودراسات المياه والتربة ومكافحة التصحر - صبراتة
[5-2] قسم الهندسة البيئية، كلية الهندسة، جامعة صبراتة، ليبيا.
[6] قسم الأحياء، كلية التربية، جامعة بني وليد، ليبيا.

Received: 20-01-2026; Accepted: 25-03-2026; Published: 10-04-2026

Abstract:

Background: Antimicrobial resistance (AMR) is a growing global public health crisis, with hospital wastewater increasingly recognized as a critical environmental reservoir of antibiotic-resistant bacteria.

Objective: This study aimed to assess the microbiological and physicochemical characteristics of hospital wastewater from Sabratha Teaching Hospital, Libya, and to evaluate their association with antibiotic resistance patterns.

Methods: A descriptive analytical laboratory-based study was conducted between June and August 2025. Wastewater samples were collected from three major discharge points within the hospital sewage system. Bacterial isolation and identification were performed using standard microbiological techniques. Antibiotic susceptibility testing was carried out according to Clinical and Laboratory Standards Institute (CLSI) and European Committee on Antimicrobial Susceptibility Testing (EUCAST) guidelines. Physicochemical parameters, including pH, total dissolved solids (TDS), biochemical oxygen demand (BOD), chemical oxygen demand (COD), and dissolved oxygen (DO), were analyzed. Statistical analysis included descriptive statistics, correlation analysis, regression modeling, and time-series forecasting.

Results: A total of 62 bacterial isolates representing six clinically significant genera were identified, with *Escherichia coli* being the predominant species. High levels of multidrug resistance (MDR) were observed, particularly among Pseudomonas spp. and *Acinetobacter spp.*, with elevated Multiple Antibiotic Resistance Index (MARI) values in samples from intensive care and emergency units. Significant discrepancies were observed between CLSI and EUCAST interpretations. A strong positive correlation was found between COD and BOD, indicating high organic pollution. COD was negatively associated with antibiotic susceptibility, whereas DO showed a positive correlation. Multivariate analysis identified COD as the strongest predictor of resistance patterns.

Conclusion: Hospital wastewater at Sabratha Teaching Hospital represents a significant hotspot for antibiotic-resistant bacteria. Both microbiological and physicochemical factors contribute to resistance dynamics. These findings highlight the urgent need for wastewater treatment systems, continuous environmental surveillance, and integrated antimicrobial stewardship strategies to mitigate ecological and public health risks.

Libyan Open University Journal of Medical Sciences and Sustainability
(https://oujournals.ly/index.php/LOUJMSS/index)
Volume 2 - Issue 1 - 2026 - Pages 38-48

Keywords: Antimicrobial resistance; hospital wastewater; multidrug-resistant bacteria; MARI; physicochemical parameters; CLSI; EUCAST; Libya

الملخص:
الخلفية: تُعد مقاومة مضادات الميكروبات (AMR) أزمة صحية عامة متزايدة على مستوى العالم، حيث يُعترف بشكل متزايد بمياه الصرف الصحي في المستشفيات كمستودع بيئي هام للبكتيريا المقاومة للمضادات الحيوية.
الهدف: هدفت هذه الدراسة إلى تقييم الخصائص الميكروبيولوجية والفيزيائية-الكيميائية لمياه الصرف الصحي في مستشفى صبراتة التعليمي، ليبيا، ودراسة ارتباطها بأنماط مقاومة المضادات الحيوية.
المنهجية: تم إجراء دراسة تحليلية وصفية مخبرية خلال الفترة من يونيو إلى أغسطس 2025. جُمعت عينات مياه الصرف من ثلاث نقاط تصريف رئيسية ضمن نظام الصرف بالمستشفى. تم عزل وتحديد الأنواع البكتيرية باستخدام التقنيات الميكروبيولوجية القياسية. أُجري اختبار حساسية المضادات الحيوية وفقًا لإرشادات معهد المعايير السريرية والمخبرية (CLSI)واللجنة الأوروبية لاختبار حساسية مضادات الميكروبات (EUCAST). كما تم تحليل الخصائص الفيزيائية-الكيميائية، بما في ذلك الرقم الهيدروجيني(pH) ، والمواد الصلبة الذائبة الكلية(TDS) ، والطلب الحيوي على الأكسجين (BOD)، والطلب الكيميائي على الأكسجين(COD) ، والأكسجين المذاب (DO). شمل التحليل الإحصائي الإحصاء الوصفي، وتحليل الارتباط، ونمذجة الانحدار، والتنبؤ بالسلاسل الزمنية.
النتائج: تم تحديد ما مجموعه 62 عزلة بكتيرية تمثل ستة أجناس ذات أهمية سريرية، وكانت الإشريكية القولونية (*Escherichia coli*)هي الأكثر شيوعًا. لوحظت مستويات مرتفعة من المقاومة المتعددة للمضادات الحيوية(MDR) ، خاصة لدى أنواع .*Pseudomonas spp*و*Acinetobacter spp.*، مع ارتفاع قيم مؤشر المقاومة المتعددة (MARI) في العينات المأخوذة من وحدات العناية المركزة والطوارئ. كما وُجدت اختلافات ملحوظة بين تفسيرات CLSI و EUCAST. أظهر التحليل وجود ارتباط إيجابي قوي بين COD وBOD، مما يشير إلى ارتفاع التلوث العضوي. كما ارتبط COD سلبًا بحساسية المضادات الحيوية، في حين أظهر DO ارتباطًا إيجابيًا. وأظهرت التحليلات متعددة المتغيرات أن COD هو أقوى مؤشر متنبئ بأنماط المقاومة.
الاستنتاج: تمثل مياه الصرف الصحي في مستشفى صبراتة التعليمي بؤرة هامة للبكتيريا المقاومة للمضادات الحيوية. وتلعب العوامل الميكروبيولوجية والفيزيائية-الكيميائية دورًا مشتركًا في ديناميكيات المقاومة. وتؤكد هذه النتائج الحاجة الملحة إلى تطبيق أنظمة فعالة لمعالجة مياه الصرف، والمراقبة البيئية المستمرة، واستراتيجيات متكاملة للحد من مقاومة مضادات الميكروبات، بهدف تقليل المخاطر البيئية والصحية العامة.

الكلمات المفتاحية: مقاومة مضادات الميكروبات؛ مياه الصرف الصحي في المستشفيات؛ البكتيريا متعددة المقاومة؛ مؤشر MARI؛ الخصائص الفيزيائية-الكيميائية؛ CLSI؛ EUCAST؛ ليبيا.

Introduction

Water pollution represents a major environmental and public health concern, particularly in coastal regions where untreated effluents can directly impact marine ecosystems and human populations. Hospital wastewater is a complex effluent containing organic matter, chemical contaminants, disinfectants, pharmaceutical residues, and pathogenic microorganisms, including antibiotic-resistant bacteria (ARB) and antibiotic resistance genes (ARGs) (1,2). These components create selective environmental conditions that facilitate the emergence, persistence, and dissemination of antimicrobial resistance (AMR) in aquatic systems (3,4).

Physicochemical parameters such as pH, total dissolved solids (TDS), biochemical oxygen demand (BOD), chemical oxygen demand (COD), and dissolved oxygen (DO) play a crucial role in shaping microbial communities and influencing bacterial survival and resistance dynamics. Elevated levels of BOD and COD indicate high organic loads, which enhance microbial proliferation and promote horizontal gene transfer, thereby accelerating the spread of antibiotic resistance (5,6). Furthermore, reduced dissolved oxygen levels are associated with increased environmental stress, which may favor the selection of resistant microbial populations (7).

Hospital wastewater has been increasingly recognized as a significant environmental reservoir of multidrug-resistant (MDR) bacteria, including clinically important species such as *Escherichia coli*, *Pseudomonas aeruginosa*, and *Acinetobacter baumannii* (8,9). The discharge of untreated or inadequately treated hospital effluents into municipal sewage systems or natural water bodies contributes to the contamination of surface and coastal waters, posing serious ecological and public health risks) 10.(

In many developing countries, including Libya, wastewater management systems are often insufficient or absent, leading to the direct release of untreated hospital wastewater into the environment. This issue is particularly critical in coastal cities such as Sabratha, where marine ecosystems and fisheries may be directly affected. Despite the growing global concern regarding environmental AMR, limited studies have investigated the combined microbiological and physicochemical characteristics of hospital wastewater in Libya and their role in driving antibiotic resistance.(11)

Therefore, this study aims to evaluate the microbiological profile and physicochemical properties of hospital wastewater from Sabratha Teaching Hospital and to investigate their association with antibiotic resistance patterns.

Problem Statement

Sabratha Teaching Hospital currently lacks an effective wastewater treatment system, resulting in the discharge of untreated effluents into the surrounding environment. Given the hospital's proximity to the Mediterranean coast, this practice poses a significant risk to marine ecosystems and public health.

Preliminary analysis of wastewater samples revealed elevated levels of organic pollution, particularly in terms of COD and BOD. The strong correlation between these parameters suggests environmental conditions that favor microbial proliferation and adaptive resistance mechanisms.

Statistical analysis demonstrated that COD is negatively associated with bacterial susceptibility to antibiotics, while DO exhibits a positive relationship with susceptibility. Multivariate regression identified COD as the most influential predictor of antibiotic resistance patterns, whereas DO emerged as a protective environmental factor.

These findings indicate that antibiotic resistance in hospital wastewater is not solely driven by antimicrobial residues but is also significantly influenced by environmental and chemical stressors.

Significance of the Study

This study is significant because it addresses the growing global concern of antimicrobial resistance (AMR) from an environmental perspective, particularly within hospital wastewater systems. While much attention has been given to clinical infections, less focus has been placed on the role of hospital effluents as reservoirs and transmission pathways for multidrug-resistant bacteria.

Study Aim

This study aims to evaluate the microbiological and physicochemical characteristics of hospital wastewater at Sabratha Teaching Hospital and to investigate their association with antibiotic resistance patterns.

Materials and Methods

Study Design

An environmental analytical cross-sectional study was conducted to evaluate the physicochemical characteristics of hospital wastewater and their association with antibiotic resistance patterns among bacterial isolates. The study integrates environmental monitoring with microbiological analysis to provide a comprehensive assessment of resistance dynamics.

Study Area and Sampling Strategy

Wastewater samples were collected from Sabratha Teaching Hospital, a major tertiary healthcare facility located in a coastal region of western Libya. Sampling was conducted over a three-month period (June–August 2025) to capture temporal variability.

Three fixed sampling points representing key discharge sources were selected:

I. Intensive Care Unit (ICU) outlet
II. Emergency department outlet
III. Main hospital wastewater discharge point

Grab sampling was performed using sterile high-density polyethylene (HDPE) containers. Samples were transported in insulated cool boxes at approximately 4°C and processed within 4–6 hours to preserve physicochemical and microbiological integrity.

Physicochemical Analysis

Physicochemical parameters were determined according to standard methods for water and wastewater analysis (APHA, 23rd edition).

The following parameters were measured:

I. pH using a calibrated digital pH meter
II. Electrical conductivity (EC) using a conductivity meter
III. Total dissolved solids (TDS) using a TDS meter
IV. Biochemical oxygen demand (BOD_5) using the 5-day incubation method
V. Chemical oxygen demand (COD) using the dichromate reflux method
VI. Dissolved oxygen (DO) using a portable DO meter

All measurements were conducted in triplicate, and mean values were reported to ensure precision and reproducibility.

Microbiological and Antibiotic Susceptibility Analysis

Bacterial isolation and identification were performed using standard microbiological techniques. Antibiotic susceptibility testing was conducted using the Kirby–Bauer disk diffusion method in accordance with guidelines from the Clinical and Laboratory Standards Institute and the European Committee on Antimicrobial Susceptibility Testing.

Multidrug resistance (MDR) was defined as resistance to three or more classes of antibiotics. The Multiple Antibiotic Resistance Index (MARI) was calculated to assess the level of resistance exposure.

Quality Assurance and Quality Control (QA/QC)

To ensure data reliability and accuracy, strict quality control measures were implemented:

I. Calibration of all analytical instruments prior to measurements
II. Use of reagent blanks and standard reference solutions
III. Triplicate analysis of all samples
IV. Use of control bacterial strains for antibiotic susceptibility testing
V. Adherence to standardized laboratory protocols

Data Integration and Statistical Analysis

Physicochemical and microbiological datasets were integrated to explore associations between environmental factors and antibiotic resistance.

Statistical analyses were performed using IBM SPSS Statistics and R software. The following analyses were conducted:

I. Descriptive statistics (mean, standard deviation)
II. Pearson correlation analysis
III. Principal Component Analysis (PCA)
IV. Canonical Correspondence Analysis (CCA)
V. Random Forest modeling to identify key predictors of resistance

Libyan Open University Journal of Medical Sciences and Sustainability
(https://oujournals.ly/index.php/LOUJMSS/index)
Volume 2 - Issue 1 - 2026 - Pages 38-48

VI. Model performance was evaluated using cross-validation techniques. A p-value < 0.05 was considered statistically significant.

Ethical and Environmental Considerations

Although the study did not involve human or animal subjects, it adhered to environmental safety and biosafety guidelines for handling wastewater and potentially pathogenic microorganisms. All laboratory procedures were conducted in accordance with institutional biosafety standards to minimize environmental contamination and researcher exposure.

Environmental and Public Health Relevance

This integrated methodological framework enabled the assessment of environmental contamination and its implications for public health. The study specifically evaluates the potential risks associated with the discharge of untreated hospital wastewater into coastal ecosystems and its role in promoting antimicrobial resistance dissemination.

Results and Discussion

Microbiological Profile of Hospital Wastewater

A total of 71 primary bacterial isolates were initially recovered from hospital wastewater samples collected at Sabratha Teaching Hospital. After data validation and quality control procedures, 62 non-duplicate and fully characterized isolates were retained for subsequent statistical analysis, representing six clinically significant bacterial genera (Figure 1). The remaining isolates were excluded due to duplication or incomplete characterization.

Escherichia coli was the predominant bacterial species, accounting for 50.0% of the isolates, followed by Acinetobacter spp., Enterobacter spp., Pseudomonas spp., and Staphylococcus spp., each representing 14.5%, while Klebsiella spp. accounted for 6.5% of the total isolates.

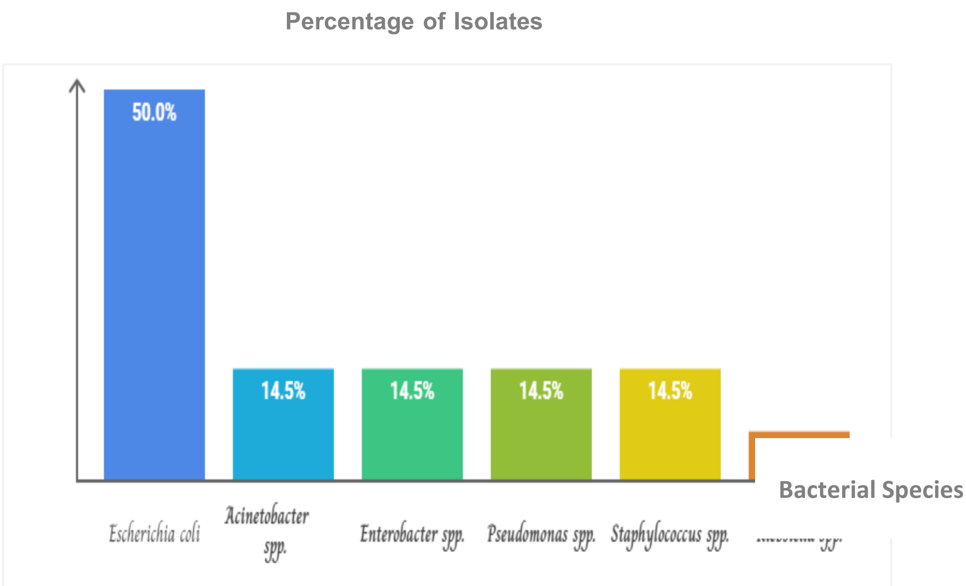


Figure 1. Distribution of Bacterial Species in Hospital Wastewater

The predominance of *Escherichia coli* reflects fecal contamination and is consistent with its role as a key indicator organism in wastewater systems. In contrast, the presence of opportunistic pathogens such as *Pseudomonas aeruginosa* and *Acinetobacter baumannii* highlights the clinical origin of the effluents. Notably, higher bacterial loads were consistently

Libyan Open University Journal of Medical Sciences and Sustainability
(https://oujournals.ly/index.php/LOUJMSS/index)
Volume 2 - Issue 1 - 2026 - Pages 38-48

observed in samples collected from the Intensive Care Unit (ICU) and Emergency Department, suggesting increased microbial burden and selective pressure in these high-risk hospital units. *Escherichia coli* Antibiotic Susceptibility.

Antibiotic Susceptibility Patterns

Antibiotic susceptibility testing was performed against a panel of approximately 20 antibiotics representing major therapeutic classes.

For *Escherichia coli* isolates, the mean inhibition zone diameter was 23.5 ± 5.2 mm, with a coefficient of variation of 22.1%, indicating moderate variability in susceptibility. A high proportion of isolates (80.6%) were classified as sensitive, while 16.1% exhibited resistance. The mean susceptibility score (0.78 ± 0.32) demonstrated a slightly negatively skewed distribution (skewness = −0.42), reflecting generally preserved susceptibility with limited extreme resistance phenotypes (**Figure** 2).
Across all bacterial genera, the overall mean susceptibility score (0.70 ± 0.36) and high coefficient of variation (51.4%) indicate substantial interspecies heterogeneity. The Shapiro–Wilk test (p = 0.06) confirmed approximate normality, supporting the validity of parametric analyses.
These findings suggest that while *Escherichia coli* maintains relatively high susceptibility levels, other species contribute disproportionately to the overall resistance burden.

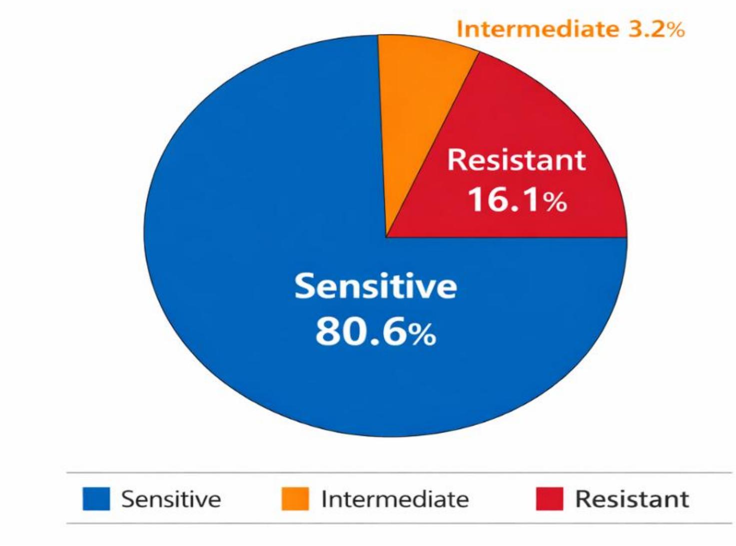


Figure 2. Antibiotic Susceptibility profile of *Escherichia coli* Isolates

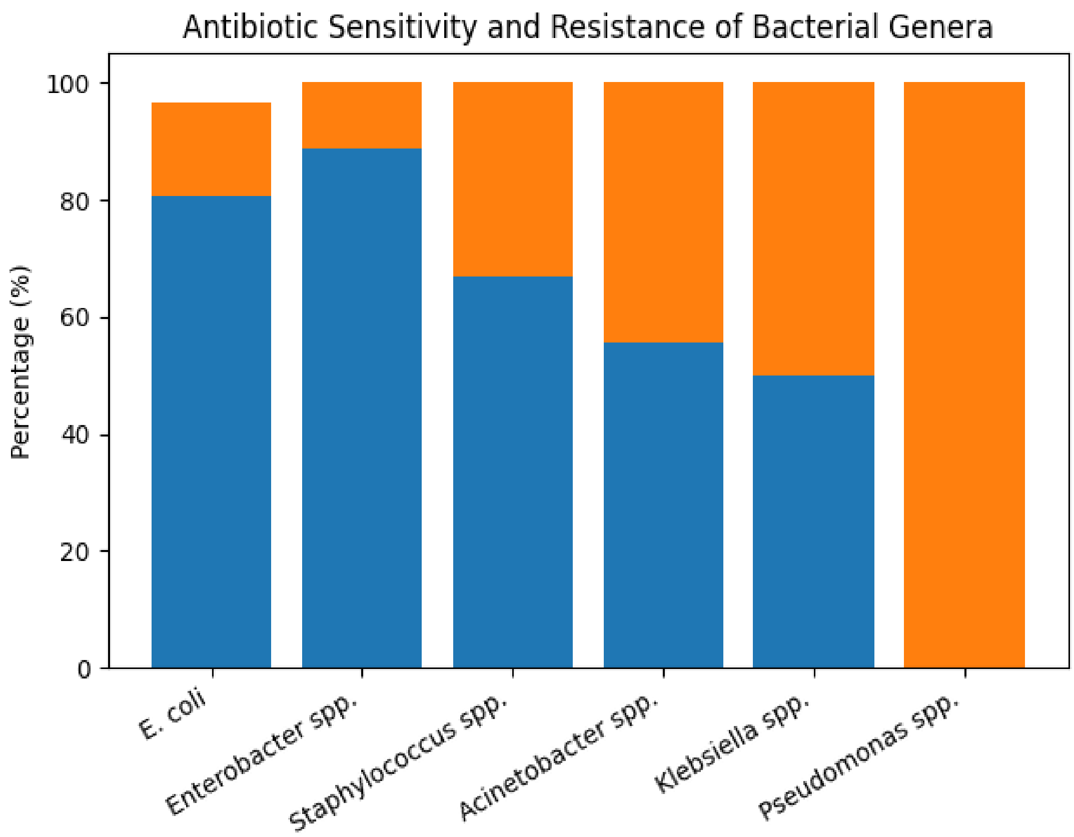
Multiple Antibiotic Resistance Index (MARI)

The weighted Multiple Antibiotic Resistance Index (MARI) for *Escherichia coli* (0.16) remained below the high-risk threshold (0.20), indicating moderate antibiotic exposure. However, isolates derived from ICU and Emergency Department effluents exhibited higher localized resistance levels, reflecting intensified selective pressure due to antibiotic usage and disinfectant exposure.
Marked interspecies variation in resistance profiles was observed (χ^2 test, $p < 0.05$).
Notably, Pseudomonas spp. exhibited complete resistance (100%; MARI = 1.00), indicating extreme multidrug resistance and strong environmental adaptation. Conversely, *Enterobacter spp.* demonstrated the highest susceptibility (88.9%; MARI = 0.11).

Libyan Open University Journal of Medical Sciences and Sustainability
(https://oujournals.ly/index.php/LOUJMSS/index)
Volume 2 - Issue 1 - 2026 - Pages 38-48

Intermediate resistance patterns were observed in Staphylococcus spp. and *Acinetobacter spp*., with resistance rates of 33.3% and 44.4%, respectively. The high variability within these groups (CV > 70%) suggests heterogeneous resistance mechanisms influenced by environmental and genetic factors. Antibiotic Sensitivity and Resistance of Bacterial Genera, **(Figure 3).**

Figure 2. Antibiotic Sensitivity and Resistance of Bacterial Genera



Influence of Physicochemical Parameters on Antibiotic Resistance

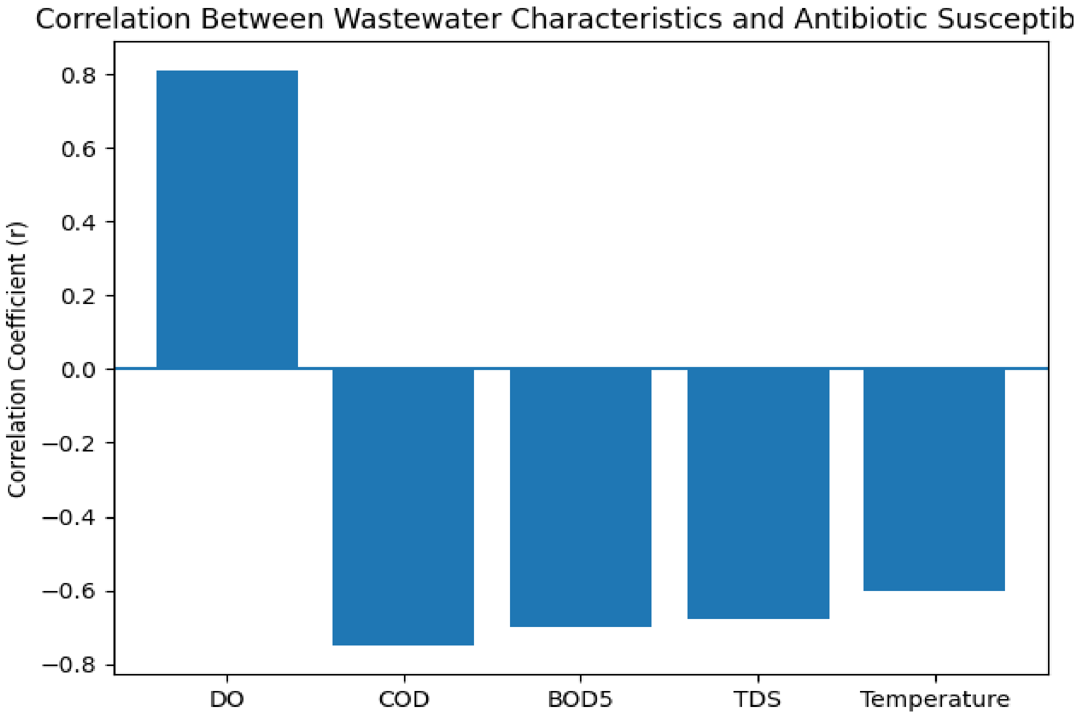
Correlation analysis revealed strong and statistically significant associations between wastewater chemical characteristics and bacterial susceptibility patterns (Figure 3).

Dissolved oxygen (DO) showed a strong positive correlation with antibiotic susceptibility (Pearson r = 0.81), indicating that higher oxygen levels may limit the proliferation of resistant strains. In contrast, key pollution indicators—COD, BOD_5, and TDS—exhibited strong negative correlations with susceptibility (r ranging from −0.68 to −0.77), suggesting that elevated organic and chemical loads promote resistance development.

Temperature also demonstrated a negative association with susceptibility, reflecting its role in enhancing microbial metabolic activity and resistance expression.

Libyan Open University Journal of Medical Sciences and Sustainability
(https://oujournals.ly/index.php/LOUJMSS/index)
Volume 2 - Issue 1 - 2026 - Pages 38-48

Species-specific interactions further supported these findings. For instance, a strong inverse relationship was observed between COD levels and ciprofloxacin susceptibility in *Escherichia coli*, while TDS showed a pronounced negative correlation with imipenem susceptibility in Pseudomonas spp., emphasizing the role of environmental stressors in shaping antibiotic resistance. Figure 4. Correlation between Wastewater Characteristics and Antibiotic Susceptible



Discussion

The present study provides a comprehensive assessment of hospital wastewater as a reservoir of antibiotic-resistant bacteria in a coastal healthcare setting. The predominance of *Escherichia coli* among the isolates is consistent with its well-established role as an indicator of fecal contamination and its widespread occurrence in wastewater systems (1,2). Similar findings have been reported in hospital effluents worldwide, where *Escherichia coli* is frequently identified as the dominant species due to its high adaptability and persistence in polluted environments (13,14).

The detection of opportunistic and clinically significant pathogens such as *Pseudomonas aeruginosa* and *Acinetobacter baumannii* further highlights the clinical origin of the wastewater and its potential role in disseminating multidrug-resistant (MDR) organisms into the environment. These species are well known for their intrinsic resistance mechanisms and ability to survive under harsh environmental conditions, including exposure to disinfectants and antibiotics (15,16).

The high bacterial loads observed in effluents from the ICU and Emergency Department are in agreement with previous studies indicating that high-risk hospital units are major contributors to antimicrobial resistance due to intensive antibiotic use and increased patient turnover (5). This supports the hypothesis that hospital wastewater is not a passive byproduct but an active hotspot for resistance selection and amplification.(17)

Antibiotic susceptibility analysis revealed that *Escherichia coli* isolates retained relatively high sensitivity levels compared to other bacterial genera. This observation aligns with reports suggesting that although *Escherichia coli* is commonly exposed to antibiotics, it may exhibit variable resistance patterns depending on environmental and clinical factors (18). In contrast, the complete resistance observed in Pseudomonas spp. (MARI = 1.00) is particularly concerning and reflects the remarkable capacity of this genus to develop multidrug resistance through mechanisms such as efflux pumps, biofilm formation, and horizontal gene transfer (19). Similarly, the elevated resistance levels in *Acinetobacter spp*. and *Klebsiella spp*. are consistent with global reports identifying these organisms as critical priority pathogens due to their resistance to multiple antibiotic classes, including carbapenems (20). The high variability observed in resistance patterns among these species suggests the influence of diverse environmental stressors and genetic adaptations.

The analysis of the Multiple Antibiotic Resistance Index (MARI) provided additional insights into the level of antibiotic exposure in the studied environment. While the overall MARI value for *Escherichia coli* remained below the high-risk threshold, localized increases in MARI values in ICU and Emergency samples indicate areas of intensified selective pressure. This finding is supported by previous studies demonstrating that MARI values above 0.20 are typically associated with high-risk contamination sources (9).

A key strength of this study lies in the integration of physicochemical parameters with microbiological data. The strong negative correlations observed between COD, BOD_5, TDS, and antibiotic susceptibility suggest that high organic and chemical loads create favorable conditions for the persistence and proliferation of resistant bacteria. These findings are in line with previous research indicating that polluted environments enhance microbial density and facilitate horizontal gene transfer, thereby accelerating the spread of resistance genes (10,11).

Conversely, the strong positive correlation between dissolved oxygen (DO) and bacterial susceptibility highlights the potential role of oxygen availability in limiting resistance development. Higher DO levels may reduce anaerobic stress conditions that favor resistant strains, thereby contributing to improved microbial susceptibility profiles (12).

Species-specific correlations further emphasize the complex interplay between environmental factors and resistance mechanisms. For instance, the inverse relationship between COD and ciprofloxacin susceptibility in *Escherichia coli*, as well as between TDS and imipenem susceptibility in Pseudomonas spp., suggests that specific environmental stressors may differentially affect antibiotic efficacy depending on the bacterial species.

The variability and outlier analysis revealed the presence of heterogeneous resistance phenotypes, which may reflect adaptive responses to fluctuating environmental conditions. The substantial agreement observed in repeated measurements ($\kappa = 0.72$) confirms the reliability of the findings while highlighting the dynamic nature of resistance expression.

Temporal analysis using ARIMA modeling suggested a potential increase in resistance levels over time, although the observed trend was not statistically significant. This projection is consistent with global concerns regarding the continuous rise of antimicrobial resistance, particularly in environments with inadequate wastewater treatment (3).

Overall, the findings of this study reinforce the concept that hospital wastewater serves as a critical environmental reservoir and transmission pathway for antibiotic-resistant bacteria. The combined influence of microbiological and physicochemical factors underscores the need for integrated management strategies, including effective wastewater treatment, environmental monitoring, and antimicrobial stewardship.

Conclusion

This study demonstrates that hospital wastewater from Sabratha Teaching Hospital represents a significant environmental reservoir of antibiotic-resistant bacteria. The predominance of clinically important species, including *Escherichia coli*, Pseudomonas spp., *Acinetobacter spp*., and *Klebsiella spp*., highlights the strong clinical origin of the detected contamination and the potential risk of dissemination into surrounding environments.

The observed high levels of multidrug resistance, particularly in Pseudomonas spp. and *Acinetobacter spp.*, together with variable MARI values across sampling sites, indicate ongoing selective pressure likely driven by antibiotic usage and inadequate wastewater management practices. In addition, significant associations between physicochemical parameters and antibiotic susceptibility—especially the strong effects of COD, BOD_5, and dissolved oxygen- confirm that environmental conditions play a crucial role in shaping resistance dynamics.

Overall, the findings emphasize that antimicrobial resistance in hospital wastewater is a multifactorial phenomenon influenced by both microbiological and environmental factors. These results highlight the urgent need for effective hospital wastewater treatment systems, routine environmental surveillance, and strengthened antimicrobial stewardship programs to reduce the dissemination of resistant bacteria and protect public and environmental health, particularly in coastal regions.

Recommendations

Based on the findings of this study, the following recommendations are proposed to reduce the environmental dissemination of antibiotic-resistant bacteria and improve hospital wastewater management in coastal healthcare settings:

1. Implementation of Wastewater Treatment Systems: It is strongly recommended that Sabratha Teaching Hospital install an effective on-site wastewater treatment system, such as membrane filtration, chlorination, or advanced oxidation processes, to reduce microbial and chemical contamination before discharge.
2. Routine Environmental Surveillance: Regular monitoring of hospital effluents should be established to track physicochemical parameters (COD, BOD_5, TDS, DO) and the presence of antibiotic-resistant bacteria, ensuring early detection of potential public health risks.

References

1. Karkman A, Do TT, Walsh F, Virta MPJ (2020). Antibiotic resistance genes in wastewater: Occurrence and dissemination. *Trends in Microbiology*.
2. Rizzo L, Manaia C, Merlin C, Schwartz T, Dagot C, Ploy MC, et al. (2019). Urban wastewater treatment plants as hotspots for antibiotic-resistant bacteria and genes. *Science of the Total Environment*.
3. World Health Organization (WHO) (2023). Global antimicrobial resistance and use surveillance report. Geneva: WHO.
4. Tacconelli E, Carrara E, Savoldi A, Harbarth S, Mendelson M, Monnet DL, et al. (2018). Discovery, research, and development of new antibiotics: The WHO priority pathogens list. *The Lancet Infectious Diseases*.
5. Hocquet D, Muller A, Bertrand X (2021). What happens in hospitals does not stay in hospitals: Antibiotic-resistant bacteria in hospital wastewater. *Clinical Microbiology and Infection*.
6. Asanousi Lamma, O., Swamy, A. V. V. S., & Alhadad, A. A. (2018). Assessment of Heavy Metal Pollution in Ground Water and its Correlation with other Physical Parameters at Selected Industrial Areas of Guntur, AP, India. AP, India.
7. Dr. Amna Ali Alhadad. (2022). Parasitic Worms: A Threat to the Global World and Economy . Bani Waleed University Journal of Humanities and Applied Sciences, 7(2), 210-228. https://doi.org/10.58916/jhas.v7i2.533
8. Zhang Y, Marrs CF, Simon C, Xi C (2020). Wastewater treatment contributes to selective increase of antibiotic resistance among bacteria. *Water Research*.
9. Pang Z, Raudonis R, Glick BR, Lin TJ, Cheng Z (2019). Antibiotic resistance in *Pseudomonas aeruginosa*: Mechanisms and alternative therapeutic strategies. *Frontiers in Microbiology*.

10. Rice LB (2020). Progress and challenges in implementing the research on multidrug-resistant organisms. *Infectious Disease Clinics of North America*.
11. Krumperman PH (1983). Multiple antibiotic resistance indexing of *Escherichia coli* to identify high-risk sources of fecal contamination. *Applied and Environmental Microbiology*.
12. Aloraibi, A. M., Alhadad, A. A., & Hamed, S. G. (2025). Antibacterial properties of alcoholic and aqueous extracts of ginger and anise plants against various types of bacteria. Bani Waleed University Journal of Humanities and Applied Sciences, 10(4), 201-212.
13. Manaia CM, Rocha J, Scaccia N, Marano R, Radu E, Biancullo F, et al. (2022). Antibiotic resistance in wastewater treatment plants: Tackling the black box. *Environment International*.
14. Guo J, Li J, Chen H, Bond PL, Yuan Z (2021). Metagenomic analysis reveals wastewater treatment plants as hotspots of antibiotic resistance genes and their dissemination. *Science of the Total Environment*.
15. Li J, Cheng W, Xu L, Jiao Y, Baig SA, Chen H (2021). The role of dissolved oxygen in shaping antibiotic resistance in aquatic environments. *Water Research*.
16. Aloraibi, A. M., Alhadad, A. A., & Ali, E. A. (2025). Isolation and Identification of Bacteria Causing UrinaryTract Infections and Their Antibiotic Sensitivity Pattern at Mohammad Almoghariaf Hospital in Ajdabiya. Libyan Open University Journal of Applied Sciences (LOUJAS), 08-18.
17. Asma M. Aloraibi, Amna Ali Alhadad, Amal Fitouri Belgasim, & Amira Mosbah Issa. (2025). Antibacterial activity of cold and hot aqueous Extract of Mentha piperita. Libyan Journal of Contemporary Academic Studies, 3(2), 52-60. https://doi.org/10.65417/ljcas.v3i2.214
18. SALEM, M. O. A., & Alhadad, A. A. (2026). Phytochemical profiling and in vitro antimicrobial potential of Hypericum decaisneanum Coss. aerial parts endemic to Bani Waleed, Libya. Libyan Open University Journal of Applied Sciences (LOUJAS), 97-103.
19. Alhadad , A. (2022). HABITAT LOSS AND ITS EFFECT ON THE BIODIVERSITY:
20. A REVIEW . Journal of Azzaytuna University, 44, 460-470