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Original Research Article

Retrospective Analysis of Antibiotic Resistance Patterns in Bacterial Isolates

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Abstract

Background: Antibiotic resistance has emerged as a critical global health concern, threatening the effective management of infectious diseases and increasing the risk of treatment failures. The WHO identifies a major public issue as antibiotic resistance, a health threat of our time, with hospital-acquired infections by multidrug-resistant organisms contributing to prolonged hospital stays, increased costs, and higher mortality. Local surveillance studies are essential to understand institution-specific resistance trends and guide rational prescribing practices.

Methods: From September 2022 to August 2024, the RDJMMCH Microbiology Department conducted this retrospective observational study. A total of 130–150 culture-positive clinical isolates were included, derived from blood, urine, sputum, pus, and wound swabs. Data were retrieved from laboratory records, and antibiotic Susceptibility testing followed CLSI recommendations using Kirby-Bauer disc diffusion. Results were analyzed to determine prevalence, resistance frequencies, and year-wise trends.

Results: Gram-negative organisms predominated, with E. coli (31.4%), and the most prevalent isolate is Klebsiella pneumoniae (18.6%), exhibiting high resistance to fluoroquinolones and cephalosporins, with rising carbapenem resistance. Among Gram-positive isolates, nearly half of Staphylococcus aureus were methicillin-resistant, and a small proportion of Enterococcus species showed vancomycin resistance. Overall, multidrug-resistant organisms were frequently encountered, with increasing resistance observed over the two years.

Conclusion: The findings highlight the urgent need for rational antibiotic prescribing, stringent infection control measures, and continuous surveillance at RDJMMCH. Regular updates to hospital antibiotic policy and effective antimicrobial stewardship programs are recommended to curb the escalation of resistance.

Keywords: Antibiotic Resistance, Bacterial Isolates, Retrospective Study, RDJMMCH.

Introduction

One of the biggest global health challenges today is antibiotic resistance. Antimicrobial resistance (AMR), which the WHO has repeatedly deemed a major public health issue, is making more diseases harder to prevent and treat. [1] The WHO's Global Antimicrobial Resistance and Use Surveillance System (GLASS) shows that drug-resistant bacterial infections cause longer hospital stays, higher healthcare costs, and death. According to the Centers for Disease Control and Prevention (CDC), antibiotic resistance results in millions of deaths and millions more illnesses [2]. Multidrug-resistant (MDR) bacteria are spreading rapidly, making it harder to treat common diseases and threatening decades of infectious disease management progress.

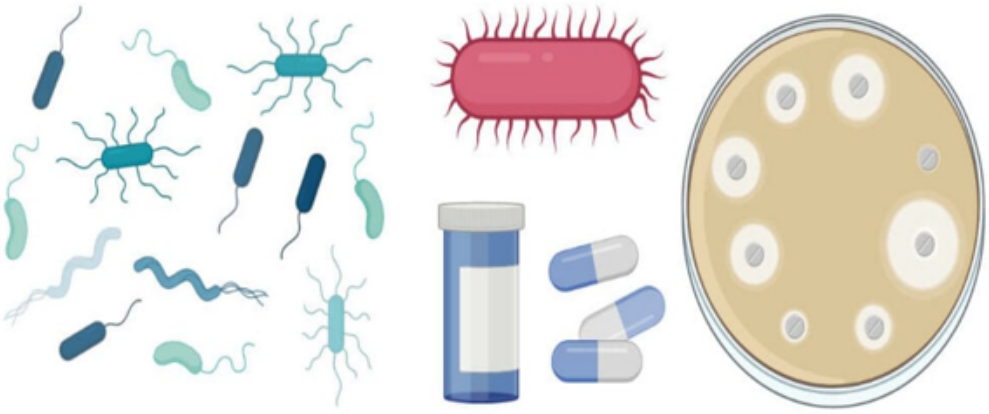

Figure 1: Antimicrobial-resistance (AMR) and Antibacterial-Resistance (ABR) [3]

India is among the low- and middle-income countries affected by antibiotic resistance [4]. Due to the country's dense population, antibiotic misuse, and weak infection control, Indians call their country the "AMR capital of the world" because of antibiotic resistance. Self-medication, antibiotics without a prescription, inadequate treatment regimens, and broad-spectrum drug misuse in community and hospital settings have exacerbated the problem [5]. Overcrowding, flawed diagnostics, and inefficient antibiotic stewardship programs aggravate hospital conditions. Bacteria and other resistant germs cause hospital-acquired infections (HAIs), which cost patients and the healthcare system time and money.

Specific multidrug-resistant organisms among resistance-linked ailments are concerning. Carbapenems, aminoglycosides, and extended-spectrum cephalosporins are ineffective against Acinetobacter baumannii, Klebsiella pneumoniae, Pseudomonas aeruginosa, and E. coli [6]. Due to the rise of extended-spectrum beta-lactamase-producing E. coli and Klebsiella bacteria, treatment options are limited. Treatment is hindered by carbapenem-resistant Enterobacteriaceae (CRE), which increases mortality [7]. Methicillin-resistant Staphylococcus aureus is a prevalent Gram-positive bacterium that can infect hospitals and the public. In immunocompromised and critical care unit patients, vancomycin-resistant Enterococci (VRE) complicate treatment decisions [8]. Due to the persistence of these diseases, local antibiogram data is essential for empirical treatment and continual surveillance.

Antibiotic resistance patterns have been studied nationally and internationally, but we need regional data to develop good local treatment strategies. Different patient demographics, infection control procedures, and prescribing practices might cause resistance patterns to vary between hospitals in the same city [9]. Institution-specific studies allow hospitals to create evidence-based antibiotic policies and stewardship programs. As a tertiary care hospital, RDJMMCH treats a wide range of patients with various medical conditions, making it suitable for resistance tracking [10]. Samples from several clinical specialisations processed by the microbiology section reveal infection prevalence and susceptibility profiles.

Multiple causes warrant this study, starting with RDJMMCH's lack of antibiotic resistance data; a systematic analysis would fill a knowledge gap. Second, using two-year data to determine if organisms' resistance is rising, stabilising, or declining is crucial to updating empirical therapy guidelines. Third, the findings will influence the hospital's antibiotic policy, encouraging more sensible prescriptions. Lastly, the data can enhance national AMR surveillance by providing a foundation for regional research and policymaking.

Aim and Objectives

This retrospective study examines antibiotic resistance profiles of RDJMMCH clinical sample bacterial isolates from September 2022 to August 2024. The specific objectives are as follows:

1. To identify bacterial isolates from clinical samples collected during the study period and determine their prevalence across different infection types.
2. Find antibiotic resistance patterns in these bacterial isolates using standard laboratory methods and classify them based on susceptibility to commonly used antimicrobials.
3. To compare resistance trends across the two-year study period (September 2022 – August 2024), identifying any notable increases or decreases in resistance rates for key pathogens.
4. To provide evidence for hospital antibiotic policy, ensuring that empirical therapy guidelines are based on local microbiological data, thus enhancing patient care and minimizing the risk of resistance escalation.

The rise of antibiotic-resistant germs is a major issue in modern medicine, especially in hospitals where susceptible persons are more prone to getting such infections. This RDJMMCH project focuses on local resistance patterns to improve infection control, clinical practice, and antimicrobial stewardship awareness. Systematic characterisation of bacterial isolates and their resistance patterns will provide a picture of the current resistance landscape and enable continuing surveillance and informed therapeutic decision-making.

Materials and Methods

Study Design: This study was retrospective observational. Because laboratory records are reliable in clinical microbiology, retrospective methods are excellent for data analysis. This study examined laboratory records over time to detect bacterial isolates and their antibiotic resistance patterns. To reduce time and money while still gaining epidemiological insights, this technique was chosen since it allows the investigation of many incidents by actively enrolling patients.

Study Setting: The research was undertaken in the microbiology department at the tertiary care hospital RDJMMCH, which diagnoses and treats a wide range of patients. The microbiological lab receives samples from medical, surgical, paediatric, gynaecological, and obstetrical departments, ICUs, and outpatient clinics. The data represent a broad population because the hospital serves urban and rural areas. The laboratory follows standard operating methods for specimen collection, culture, and sensitivity testing to ensure dependability and consistency.

Study Period: The study lasted two years, from September 2022 to August 2024. This time span was utilised to collect enough data to evaluate resistance patterns across several seasons because seasonal circumstances and annual bacterial occurrence can affect infections and resistance patterns.

Sample Size: A total of 130–150 clinical isolates were culture-positive. Throughout the investigation, complete laboratory records determined this sample size. To avoid bias and duplicate resistance frequency reporting, only one isolate per patient was considered.

Inclusion Criteria

- Bacterial isolates obtained from clinical specimens (blood, urine, sputum, pus, wound swabs, and other body fluids).
- Specimens processed by the microbiology laboratory during the study period.
- Both Gram-positive and Gram-negative organisms are included.
- Only significant pathogens considered, based on clinical and microbiological judgment.

Exclusion Criteria

- Duplicate isolates from the same patient.
- Contaminated cultures with no clinical significance.
- Incomplete laboratory records lacking essential details of bacterial identification or susceptibility results.

Data Collection: Data were retrospectively collected from computer databases and microbiological lab reports. Data included patient information, clinical specimen type, isolated bacterial species, and antibiotic susceptibility test results. Data was anonymised during the trial to protect patient privacy.

Antibiotic Susceptibility Testing: Mueller-Hinton agar's antimicrobial susceptibility was tested using the disc diffusion method (Kirby-Bauer technique) per Clinical and Laboratory Standards Institute guidelines (CLSI). Multiple isolates' resistance patterns were confirmed using the lab's automated procedures. Beta-lactams, aminoglycosides, fluoroquinolones, carbapenems, glycopeptides, and linezolid were tested, depending on the organism. CLSI breakpoints classified isolates as sensitive, intermediate, or resistant.

Data Analysis: Standard statistical methods were employed to compile and examine the data. Percentages and frequencies were calculated to show bacterial isolate distribution and resistance trends. Year-wise comparisons were made to assess resistance September 2022–August 2023 and September 2023–August 2024 trends.

The results showed bacteria and microbial resistance patterns graphically and tabularly. The study sought to identify common organisms, their antibiotic susceptibility, and any emerging multidrug-resistant strains.

Results

Demographic Profile of Cases: This study contained 140 culture-positive bacterial isolates. Patients ranged in age from 5 to 82, with a mean of 43.2. The majority of isolates were obtained from adults aged 21–60 years (62.1%), followed by elderly patients over 60 years (24.3%) and children under 20 years (13.6%). Gender distribution revealed a slightly higher prevalence among males (56.4%) compared to females (43.6%). This male predominance was observed consistently across both years of the study.

Distribution of Bacterial Isolates: Out of the 140 isolates, Gram-negative organisms constituted the majority (72.1%), while Gram-positive cocci accounted for 27.9%. Among Gram-negative

bacilli, Escherichia coli (31.4%), Klebsiella pneumoniae (18.6%), Pseudomonas aeruginosa (12.9%), and Acinetobacter baumannii (9.3%). Gram-positive agents include Staphylococcus aureus (MRSA and MSSA), which was predominant (19.3%), followed by Enterococcus species (8.6%).

Table 1: Distribution of bacterial isolates (N = 140)

Organism	Number of isolates	Percentage (%)
Escherichia coli	44	31.4
Klebsiella pneumoniae	26	18.6
Pseudomonas aeruginosa	18	12.9
Acinetobacter baumannii	13	9.3
Staphylococcus aureus	27	19.3
Enterococcus spp.	12	8.6
Total	**140**	**100**

Antibiotic Resistance Patterns

Gram-Negative Bacilli: Highly resistant Gram-negative isolates were found. E. coli and Klebsiella spp. were resistant to the third-generation cephalosporins (72.7% and 69.2%), indicating widespread ESBL production. Klebsiella and E. coli isolates were 28.6% and 22.7% imipenem/meropenem-resistant, respectively. Aminoglycosides such as amikacin retained relatively better activity, with resistance rates around 15–20%.

Fluoroquinolone resistance was widespread, particularly among urinary isolates, with nearly half of E. coli resistant to ciprofloxacin.

Table 2: Resistance patterns of Gram-negative organisms

Antibiotic class	E. coli (n=44)	Klebsiella (n=26)	Pseudomonas (n=18)	Acinetobacter (n=13)
Cephalosporins	72.7%	69.2%	55.6%	61.5%
Carbapenems	22.7%	28.6%	38.9%	46.2%
Aminoglycosides	18.2%	19.2%	33.3%	38.5%
Fluoroquinolones	47.7%	42.3%	44.4%	53.8%

Gram-Positive Cocci: Among Staphylococcus aureus isolates, methicillin resistance (MRSA) was detected in 44.4% of cases. Resistance to penicillin was universal (100%), while resistance to macrolides (erythromycin) was noted in 48.1% of isolates. Importantly, all S. aureus isolates remained sensitive to vancomycin and linezolid. Among Enterococcus spp., 16.7% showed VRE, while all were sensitive to linezolid.

Table 3: Resistance patterns of Gram-positive organisms

Antibiotic class	S. aureus (n=27)	Enterococcus spp. (n=12)
Penicillin	100%	83.3%
Macrolides	48.1%	41.7%
Vancomycin	0%	16.7%
Linezolid	0%	0%

Trends Over Two Years: When comparing resistance rates between the first year (Sept 2022 – Aug 2023) and the second year (Sept 2023 – Aug 2024), a gradual increase in resistance was observed across most Gram-negative bacilli. Notably, carbapenem resistance in Klebsiella increased from 23.1% in the first year to 34.6% in the second year.

Similarly, fluoroquinolone resistance in E. coli rose from 43.5% to 52.2% over the two years. For Gram-positive organisms, the proportion of MRSA remained relatively stable, though a slight rise in macrolide resistance was recorded. VRE prevalence also increased marginally from 12.5% to 20%. These findings suggest a worrisome upward trend in resistance, underscoring the need for continuous surveillance and stricter antibiotic stewardship.

Discussion

This study examines 140 RDJMMCH bacterial isolates' antibiotic resistance patterns over two years. Most of the bacteria were Gram-negative, with Klebsiella pneumoniae and Escherichia coli occurring most often. In line with the rise in ESBL-producing germs, these microorganisms showed increased fluoroquinolone and third-generation cephalosporin resistance. Due to carbapenems' use for serious illnesses, Klebsiella and Acinetobacter's resistance is concerning. 20% of Gram-positive

bacteria were Staphylococcus aureus, and 50% were methicillin-resistant. Knowing that all germs are vulnerable to linezolid and vancomycin is comforting, but identifying vancomycin-resistant Enterococcus is a serious therapeutic challenge.

The findings indicate that multidrug resistance is increasing, making empirical treatment harder.

Comparison with Studies from India and Global Literature: This study matches global and country antibiotic resistance data. A multicentric ICMR investigation found Klebsiella and Escherichia coli to be the most common Gram-negative bacteria resistant to fluoroquinolones and cephalosporins [11]. The current study found 22-29% carbapenem resistance in Enterobacteriaceae, up from 20-30% in prior research from tertiary care institutions in Mumbai, Delhi, and Chennai. Due to their severe resistance patterns, the WHO has identified E. coli, Klebsiella, Pseudomonas, and Acinetobacter as "critical priority pathogens" for research and treatment development [12].

The study's 44% Gram-positive MRSA prevalence is slightly higher than the global average of 30%, although it falls within the 30-50% range estimated in Indian hospitals. Even though VRE are rare, they reflect a global issue: Enterococcus vancomycin resistance in Europe and North America is rising. Our consistent findings with national and worldwide statistics demonstrate the importance of frequent local surveillance in AMR trend monitoring.

Table 4: Previous antibiotic resistance studies compared to the present study

Study	Study Type	Sample Size	Key Findings
Present Study	Retrospective observational	140 bacterial isolates	E. coli and Klebsiella predominated; high resistance to cephalosporins and fluoroquinolones; rising carbapenem resistance; 44% MRSA; few VRE cases.
Study 1 [13]	Multicentric surveillance	>5000 isolates	High prevalence of ESBL-producing E. coli and Klebsiella; carbapenem resistance 20–30%; MRSA prevalence ~40%; emerging colistin resistance noted in some centers.
Study 2 [14]	Retrospective hospital-based	320 isolates	E. coli most common isolate; cephalosporin resistance >70%; MRSA prevalence 35%; VRE detected in 12% of Enterococcus.
Study 3 [15]	Global surveillance	>40,000 isolates (multi-country)	Critical pathogens: E. coli, Klebsiella, Pseudomonas, Acinetobacter; cephalosporin resistance widespread (>60% in many countries); MRSA ~30% globally; VRE increasing in high-income countries.

Emergence of Multidrug-Resistant Organisms: MDR bacteria are increasing in this study, which is alarming. Multipdrug-resistant Klebsiella and Acinetobacter cause hospital-acquired infections, longer hospital stays, and higher mortality.

Increasing MRSA and VRE rates limit treatment options and compel expensive or dangerous replacements. Since carbapenem and fluoroquinolone resistance have been rising year after year, antibiotic selection pressure is making this a growing problem. These trends may accelerate the day when even minor infections can be lethal due to a lack of appropriate therapies.

Clinical Implications:

Empirical Therapy Challenges: Clinical practice is significantly affected by the results. Empirical therapy, which uses local resistance to choose antibiotics, is harder when resistance rates are high. Example: cephalosporins. E. coli and Klebsiella are resistant to these antibiotics; thus, they might not constitute the most effective choice for treating kidney, bladder, or bloodstream infections at RDJMMCH. The rise of carbapenem resistance casts a question on their utility in treating severe sepsis and ICU infections. Since Methicillin-Resistant Staphylococcus aureusis (MRSA) is common in Gram-positive infections, empirical coverage with anti-MRSA medications may be warranted in many clinical situations. Providing effective coverage without promoting resistance is still a challenge.

Limitations of the Study: Despite their usefulness, the results include several drawbacks. The study only covered one site; thus, the results may not apply to other places or organisations. The facility and surroundings can greatly affect resistance patterns. The retrospective study employed preexisting laboratory data, which may have missed important clinical information or outcomes. While 130–150 isolates are enough for exploratory research, they may not reflect rare organisms or resistance mechanisms. Despite these limitations, the analysis captures the RDJMMCH resistance pattern throughout the research period.

Strengths of the Study

Despite its flaws, the study has some merit. Institution-specific resistance data guides empirical therapy at RDJMMCH. The standardised susceptibility testing methods make the results more dependable, and new patterns can be found

over the two-year study period. Additionally, the study provides baseline data for future surveillance and adds to India's antibiotic resistance research. The findings can help hospitals adjust antibiotic policies and implement effective stewardship measures.

Conclusion

Our two-year retrospective investigation of 140 bacterial isolates at RDJMMCH demonstrates that antibiotic resistance is growing in tertiary care facilities. Gram-negative bacteria like Escherichia coli and Klebsiella pneumoniae predominated. The development of carbapenem resistance and fluoroquinolone and third-generation cephalosporin resistance in these bacteria is alarming. Most Gram-positive bacteria were Staphylococcus aureus, with half being methicillin-resistant. Some Enterococcus species were vancomycin-resistant. These multidrug-resistant infections threaten patient care by complicating empirical therapy decisions and limiting therapeutic options. These findings emphasise the necessity of rationally prescribing antibiotics using local antibiogram data and strictly preventing the spread of antibiotic-resistant bacteria, which can spread diseases. Resistance patterns must be monitored to identify emerging trends and guide rapid actions. To promote judicious antibiotic usage and preserve efficacy, RDJMMCH must create and update an antibiotic policy that is completely integrated with antimicrobial stewardship activities. By synchronising local and global policies, the hospital can reduce antibiotic resistance and improve patient outcomes.

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