

Microbiological Profile of Respiratory Tract Infections and Antibiotic Sensitivity Pattern: A Retrospective Study

Lt Colonel (Dr.) Gaurav Singh

Assistant Professor, Department of Microbiology, Lord Buddha Koshi Medical College & Hospital, Saharsa, Bihar, India

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Corresponding Author: Lt Colonel (Dr.) Gaurav Singh

Conflict of interest: Nil

Abstract:

Background: Infections of the respiratory tract (RTIs) are one of the most frequent reasons for illness and death on a global scale. The rise in antimicrobial resistance to respiratory bacteria necessitates the regular assessment of the microbiological spectrum and antibiotic sensitivity for correct empirical treatment.

Aim: To analyse the microbiological composition of respiratory tract infections and to identify patterns of antibiotic susceptibility in bacterial isolates obtained from respiratory samples.

Methodology: A retrospective observational study was performed in the Department of Microbiology, Lord Buddha Koshi Medical College & Hospital, Saharsa, Bihar, India, for one year. 120 respiratory samples from patients clinically diagnosed with respiratory tract infection were selected. The laboratory data were reviewed to get the information about demographic profile, type of respiratory sample, identification of isolated bacteria, and their antibiotic sensitivity profile. Microbiological techniques were used for bacterial identification, and the Kirby-Bauer disc diffusion technique, in accordance with CLSI guidelines, was employed for the antibiotic sensitivity test.

Results: The sputum sample was the highest number of respiratory samples received (50.0%). The main bacterium isolated was *Klebsiella pneumoniae* (30.0%), while *Pseudomonas aeruginosa* and *Staphylococcus aureus* were the second and third most common bacteria respectively at 20.8% and 16.7%. Meropenem (90.8%), Amikacin (86.7%), and Piperacillin-Tazobactam (82.5%) were the most susceptible to antimicrobial drugs, while Ampicillin showed the greatest drug

Conclusion: The gram-negative bacteria accounted for the leading causative agents in respiratory infections, with the most prevalent isolates being *Klebsiella pneumoniae*. The high rate of multidrug resistance and the resistance trend to frequently prescribed antibiotics underscore the need for microbiological cultures and antibiotic sensitivity testing in order to facilitate appropriate antimicrobial stewardship.

Keywords: Respiratory tract infections; *Klebsiella pneumoniae*; Antibiotic susceptibility; Antimicrobial resistance; Multidrug-resistant organisms; Retrospective study.

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Introduction

One of the leading causes of illness, hospitalisation, and death across all age groups is respiratory tract infections, which rank high among the most common infectious diseases globally. Many different types of infections can affect either the upper or lower respiratory tracts, and are collectively known as respiratory tract infections (RTIs). Although there are numerous respiratory virus infections, bacteria play a critical role in the etiology of many community-acquired and nosocomial RTIs, especially among children, elderly people, immunocompromised patients, and individuals suffering from chronic diseases. Respiratory tract infections (RTIs) are a major economic and health burden worldwide due to the wide range of symptoms they can cause, from a simple upper

respiratory infection to pneumonia, COPD acute exacerbation, and respiratory failure [1].

Despite advancements in vaccination, antibiotic treatment, and supportive care, respiratory tract infections continue to be a major concern for public health services, leading to millions of visits to outpatient clinics and hospitalizations each year. Worldwide estimates place lower respiratory tract infections among the leading infectious illness killers, particularly in underdeveloped nations. This burden is additionally increased by the urbanization, aging of the population, environmental pollution, smoking, chronic respiratory diseases, diabetes mellitus, and immunosuppression that make patients more susceptible to respiratory infections and poor prognosis [2,3].

Bacterial infections of the respiratory tract differ depending on the region, type of health-care facility, patient characteristics, season, and comorbidities. Among common bacterial pathogens, there are *S. pneumoniae*, *H. influenzae*, *S. aureus*, *K. pneumoniae*, *P. aeruginosa*, *A. baumannii*, *E. coli*, and other Gram-negative bacilli. Prevalence of these microorganisms differs in community- and hospital-acquired infections and makes the microbiological surveillance important for the empirical choice of antimicrobial therapy [4]. Understanding of the spectrum of bacterial pathogens in the institution allows clinicians to prescribe appropriate antibiotics without unnecessary broad-spectrum antimicrobial therapy [5].

The main problem in the treatment of respiratory tract infections is the rapid increase in the number of antimicrobial resistances. Overuse of antibiotics in the community and in the hospitals leads to the development of multidrug-resistant (MDR) microorganisms, enterobacteria producing ESBLs, MRSA, carbapenem-resistant Gram-negative bacilli, and other drug-resistant pathogens [6]. Antimicrobial resistance severely limits treatment options, prolongs hospital stay, increases the cost of the treatment, and results in increased morbidity and mortality [7]. Therefore, regular surveillance of antibiotic susceptibility becomes an important part of antimicrobial stewardship and evidence-based medicine [8].

Microbiological culture and antibiotic susceptibility test are the basic methods of detecting pathogen and choosing the best treatment [9]. In addition to targeted therapy, microbiological diagnosis allows to reduce the unnecessary empirical antibiotic therapy and prevents the emergence of antimicrobial resistance [10]. Evaluation of the local antibiotic susceptibility profiles allows to update the guidelines of empirical therapy regularly [11].

The retrospective hospital-based study provides epidemiologically relevant information regarding the distribution of pathogens and their susceptibility to antibiotics based on data from routine laboratory investigations [12]. This is especially helpful in detecting changes in bacterial species, emergence of resistance mechanisms, and changes in antibiotic susceptibility over time. Results obtained in such studies help in the design of preventive strategies, empirical treatment of infection, and antibiotic stewardship policies at the institution level [13].

Several studies on the respiratory pathogens from various geographical areas have been performed; however, there is variation in bacterial spectra and resistance patterns among different healthcare facilities [14]. It is imperative that facility-specific results are taken into account while formulating treatment protocol to avoid unnecessary use of antibiotics [15]. Research on the prevalence and

characteristics of respiratory tract infections in Bihar's tertiary care hospitals is scant.

Methodology

Study Design: This research study has been conducted using a retrospective observational design for the purpose of establishing the microbial profile of RTI and susceptibility pattern of the bacteria isolated from the respiratory samples. This research study has been carried out using the systematic analysis of laboratory records for identifying the frequency of the bacteria, distribution of the respiratory samples, and antimicrobial susceptibility patterns. The choice of the retrospective design is based on the fact that it would provide the means to analyze laboratory data, identify pathogen distribution, and antimicrobial resistance pattern.

Study Setting: This research was carried out at the Department of Microbiology, Lord Buddha Koshi Medical College & Hospital, Saharsa, Bihar, India.

Study Duration: The study took place over the span of one year.

Study Population: The study group included patients who presented symptoms of respiratory tract infections and had their respiratory samples cultured at the Department of Microbiology in order to test them for their bacterial composition and antibiotic susceptibility profile. Both hospitalized and ambulatory patients, regardless of their age and sex, could be included in the study group, if there was a full microbiological data for them.

Inclusion Criteria

- Patients who have been clinically diagnosed with suspected to be infected with respiratory tract infection.
- The Department of Microbiology receives respiratory samples for culture and sensitivity testing.
- Positive respiratory cultures based on the microbiological criteria of positivity.
- Both male and female patients belonging to any age group.
- Records of complete microbiological and sensitivity test results in the laboratory database.

Exclusion Criteria

- Respiratory samples without evidence of any bacterial growth.
- Duplicate cultures taken from the same individual during an infection.
- Contaminated samples taken or processed during the laboratory procedure.
- Laboratory records which do not have information on culture or antibiotic sensitivities.

- Respiratory samples with presence of normal respiratory flora only and no pathogens.

Sample Size: A total of 120 samples from suspected respiratory tract infections were collected for this study. The samples were chosen retroactively using the microbiology laboratory records of the Department of Microbiology, Lord Buddha Koshi Medical College & Hospital, Saharsa, Bihar. All samples which satisfied the inclusion criteria of the study and produced clinically significant bacteria with complete sensitivity reports were used in the study. Samples which had duplicate isolations in the same patient with the same infection episode and those with incomplete microbiology records were excluded from the study.

Respiratory Specimen Collection: Specimen collection was performed by qualified medical personnel using standard procedures. The type of specimen varied depending on the clinical presentation and included either sputum, endotracheal aspirates, BAL, tracheal secretion, pleural fluid, or other clinically relevant respiratory specimens. All specimens were delivered rapidly to the Department of Microbiology in sealed leak-proof containers and processed using standard laboratory procedures.

Microbiological Processing: Respiratory samples were studied macroscopically and microscopically prior to culturing. The appropriate materials were cultivated using standard microbiological medium, including Blood Agar, MacConkey Agar, and Chocolate Agar. We watched the cultures for bacterial development after we let them grow in the right environments. Common procedures utilised in microbiology labs, such as colony traits, Gram staining features, and biochemical testing, were utilised to identify the isolates.

Antibiotic Susceptibility Testing: In accordance with the Clinical and Laboratory Standards Institute (CLSI) recommendations, all bacterial isolates that were clinically relevant were tested for antibiotic susceptibility using the Kirby-Bauer disc diffusion technique on Mueller-Hinton agar. As part of standard operating procedure, the hospital would conduct antibiotic susceptibility testing on a variety of bacterial species to determine which antibiotics would be most effective. The test findings were categorized as S (sensitive), I (intermediate), or R (resistant) based on the most recent CLSI understanding. Quality control of susceptibility testing was done using known reference bacterial strains.

Data Collection: Retrospective data were obtained from the laboratory records and the Laboratory

Information System through a structured proforma. These data included patient's age, gender, hospitalization status, type of specimen, bacterium isolated, Gram stain results, and antibiotic susceptibility test results. Patient privacy was ensured through the complete de-identification of all patients' data before any analysis was performed.

Outcome Measures: The main variables of measurement were:

- Distribution of samples collected from the respiratory system for culture.
- Frequency of occurrence of bacterial pathogens causing respiratory tract infections.
- Distribution of Gram-positive and Gram-negative bacteria isolates.
- Patterns of antibiotic sensitivity for isolates.
- Frequency of MDR bacterial isolates.

Statistical Analysis: Microsoft Excel was used to record the data, and IBM SPSS Statistics Version 27.0 (IBM Corp., Armonk, NY, USA) was used for analysis. The purpose of the descriptive analysis was to offer a synopsis of the demographic data, specimens collected, bacteria identified, and antibiotic resistance. For categorical data, we used percentages and frequencies, while for continuous data, we used means $\pm$ SD. The Chi-square test and Fisher's exact test were used to evaluate the associations between the categorical data. A statistically significant result was defined as $P < 0.05$.

Results

A total of 120 specimens from respiratory tracts sent to the Department of Microbiology were considered for analysis in the current retrospective study. The isolated bacteria were examined in order to evaluate the demographic profile of patients, type of respiratory specimens, bacterial species isolated, staining properties on Gram staining and their antibiotic sensitivity.

Demographic Characteristics of the Study

Population: Table 1 displays the demographic information of the people who participated in the study. Respiratory tract infections were most common in middle-aged and older individuals, with the highest numbers of cases observed in the age groups of 46-60 years (35.0%) and 31-45 years (25.0%). Males accounted for 61.7% of study subjects whereas females represented 38.3% of the total population. Most of the samples were collected from hospitalized patients (58.3%), ICU patients (23.3%), and outpatients (18.3%).

Table 1: Demographic Characteristics of the Study Population		
Variable	N	Percentage (%)
Age Group (Years)		
≤15	12	10.0
16–30	18	15.0
31–45	30	25.0
46–60	42	35.0
>60	18	15.0
Gender		
Male	74	61.7
Female	46	38.3
Patient Category		
Outpatient (OPD)	22	18.3
Inpatient (Ward)	70	58.3
Intensive Care Unit (ICU)	28	23.3

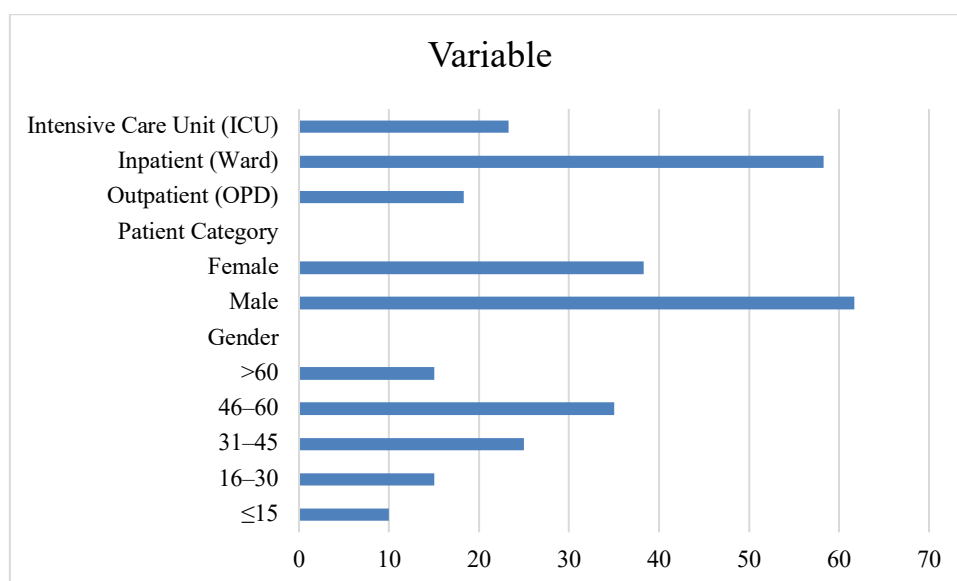


Figure 1: Graphical Representation of Demographic Characteristics of the Study Population

Distribution of Respiratory Specimens: Sputum specimens were most common, making up 50% of the total, while endotracheal aspirates comprised

20.8% of the total specimens collected. Bronchoalveolar lavage specimens comprised 13.3% of the specimens collected.

Table 2: Distribution of Respiratory Specimens Received (n = 120)		
Specimen Type	Frequency (n)	Percentage (%)
Sputum	60	50.0
Endotracheal Aspirate	25	20.8
Bronchoalveolar Lavage (BAL)	16	13.3
Tracheal Aspirate	10	8.3
Pleural Fluid	5	4.2
Others	4	3.4

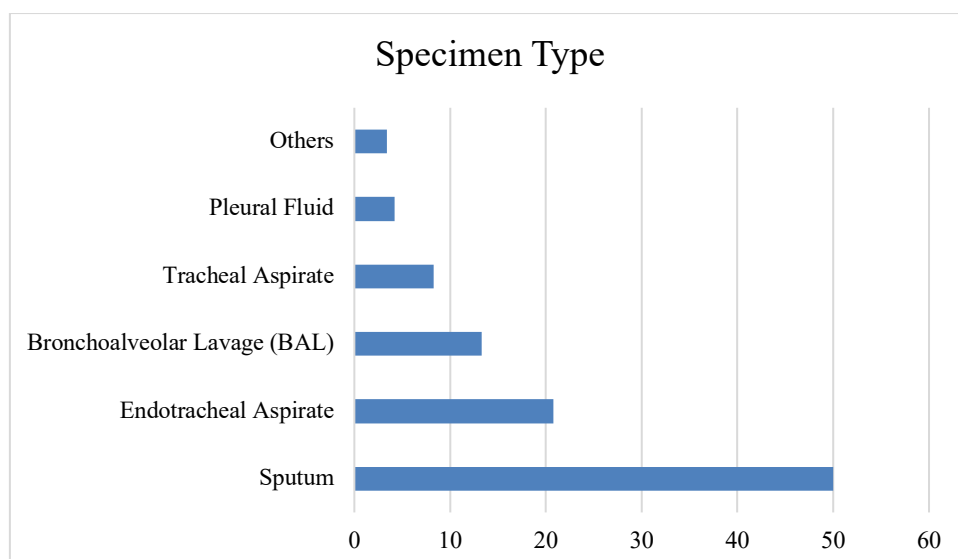


Figure 2: Graphical Representation of Distribution of Respiratory Specimens Received (n = 120)

Microbiological Profile of Bacterial Isolates:

Table 3 shows the distribution of the isolated bacteria among the respiratory samples. The predominant bacteria in the samples was *Klebsiella pneumoniae* (30.0%), followed by *Pseudomonas*

aeruginosa (20.8%), *Staphylococcus aureus* (16.7%) and *Escherichia coli* (10.8%). Other significant bacteria were *Acinetobacter baumannii*, *Streptococcus pneumoniae*.

Table 3: Distribution of Bacterial Isolates from Respiratory Specimens (n = 120)		
Bacterial Isolate	Frequency (n)	Percentage (%)
<i>Klebsiella pneumoniae</i>	36	30.0
<i>Pseudomonas aeruginosa</i>	25	20.8
<i>Staphylococcus aureus</i>	20	16.7
<i>Escherichia coli</i>	13	10.8
<i>Acinetobacter baumannii</i>	10	8.3
<i>Streptococcus pneumoniae</i>	8	6.7
<i>Haemophilus influenzae</i>	5	4.2
<i>Enterobacter</i> spp.	3	2.5
Total	120	100.0

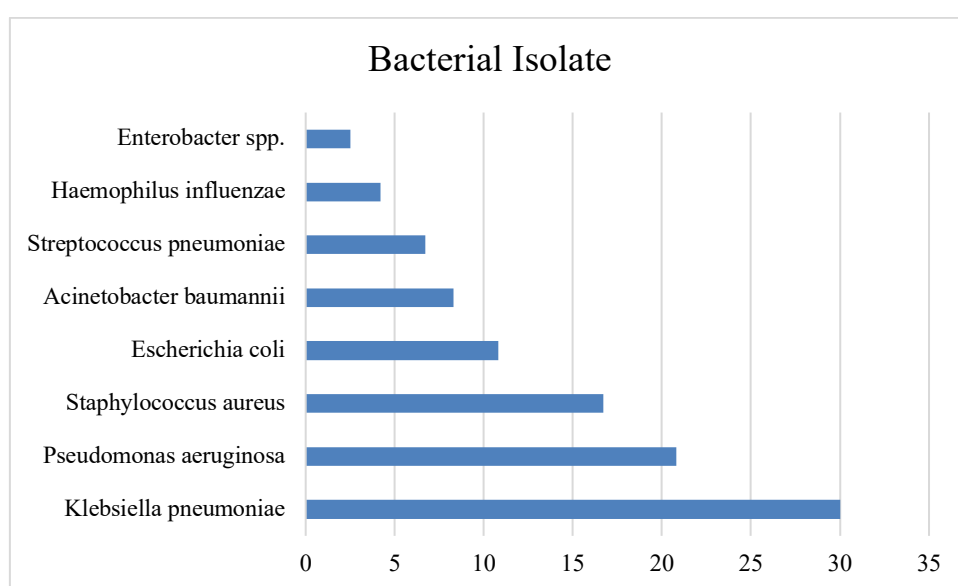


Figure 3: Graphical Representation of Distribution of Bacterial Isolates from Respiratory Specimens (n = 120)

Antibiotic Susceptibility Pattern of Major Bacterial Isolates: According to the study on the antibiotic susceptibility, Meropenem (90.8%), Amikacin (86.7%), and Piperacillin-Tazobactam

(82.5%) were the most effective antibiotics for the bacteria. The level of susceptibility of Cefepime and Ciprofloxacin was moderate while Ampicillin had the least susceptibility level of 25.8%.

Table 4: Overall Antibiotic Susceptibility Pattern of Bacterial Isolates (n = 120)

Antibiotic	Susceptible n (%)	Resistant n (%)
Ampicillin	31 (25.8)	89 (74.2)
Amoxicillin–Clavulanate	52 (43.3)	68 (56.7)
Ceftriaxone	58 (48.3)	62 (51.7)
Cefepime	75 (62.5)	45 (37.5)
Ciprofloxacin	72 (60.0)	48 (40.0)
Amikacin	104 (86.7)	16 (13.3)
Piperacillin–Tazobactam	99 (82.5)	21 (17.5)
Meropenem	109 (90.8)	11 (9.2)

Distribution of Multidrug-Resistant (MDR) Organisms: The distribution of multidrug-resistant bacteria is shown in Table 5 below. Out of the 120 isolated bacteria, 39 (32.5%) were found to be multidrug-resistant bacteria. The prevalence rate of

multidrug-resistant isolates in the three types of respiratory bacteria is highest in *K. pneumoniae* (35.9%), followed by *A. baumannii* (20.5%) and *P. aeruginosa* (17.9%).

Table 5: Distribution of Multidrug-Resistant (MDR) Bacterial Isolates (n = 39)

Organism	MDR Isolates (n)	Percentage (%)
<i>Klebsiella pneumoniae</i>	14	35.9
<i>Acinetobacter baumannii</i>	8	20.5
<i>Pseudomonas aeruginosa</i>	7	17.9
<i>Escherichia coli</i>	4	10.3
<i>Staphylococcus aureus</i> (MRSA)	4	10.3
<i>Enterobacter</i> spp.	2	5.1
Total MDR isolates	39	100.0

This study evaluated 120 respiratory samples; the most common type of sample was sputum, and the most prevalent bacterial pathogen was *Klebsiella pneumoniae*, followed by *Pseudomonas aeruginosa* and *Staphylococcus aureus*. The antibacterial activity of carbapenems and aminoglycosides was the greatest compared to other groups, while the rate of bacteria resistant to Ampicillin and Amoxicillin-Clavulanate was quite high. It was found that almost one third of all the isolates were multidrug-resistant.

Discussion

This paper from the Department of Microbiology at Lord Buddha Koshi Medical College & Hospital in Saharsa, Bihar, discusses the antibiotic susceptibility pattern of bacteria isolated from respiratory tract infections (RTIs) and the microbiological profile of RTIs. The bulk of the bacteria that were isolated were Gram-negative; the organisms that were most commonly found were *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*, according to Sadhna and Ranjana (2018) [16]. Moreover, Carbapenems and aminoglycosides were found to be the most active anti-microbial drugs, while there was significant resistance against the commonly used β -lactam group of anti-microbials.

Most cases of RTIs were seen in patients aged 46–60 years and the majority were male patients. These results were in agreement with other studies which have revealed that aging, smoking, chronic respiratory diseases, diabetes mellitus, and other comorbid conditions make the patients prone to bacterial respiratory infection Samad et al. (2017) [17]. The sputum samples comprised the largest proportion of samples received in the laboratory followed by the endotracheal aspirates and bronchoalveolar lavage specimens, which was in line with reports from tertiary care hospitals where sputum continues to be the principal sample for the diagnosis of lower respiratory tract infections.

Klebsiella pneumoniae, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Escherichia coli* were the most common bacteria isolated in our study. This confirms previous research showing that Gram-negative bacilli are common in healthcare facilities and can cause respiratory tract infections, particularly in very sick patients. Researchers Singh et al. (2020) [18]. The prevalence of these isolates has been attributed to their ability to produce multiple virulence factors and increased antimicrobial resistance.

Antimicrobial efficacy against the identified pathogens was best demonstrated by sensitivity testing with Meropenem, Amikacin, and Piperacillin-Tazobactam. Past research in India and elsewhere has shown that carbapenems and aminoglycosides are effective against severe respiratory infections; our microorganisms' sensitivity pattern is consistent with this. On the other hand, Sodani and Hawaldar (2020) [19] noted that the rising issue of anti-microbial resistance, which is caused by the misuse of these drugs and ESBL producing organisms, was demonstrated by resistance to Amoxicillin-clavulanate, third generation cephalosporins, and ampicillin.

Almost one-third of the isolates in the current study were multidrug resistant (MDR). The highest frequency was recorded for *Klebsiella pneumoniae* followed by *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. Similar reports are available throughout the world indicating the rising trend of MDR respiratory pathogens and the need for continuous monitoring through microbiology, prescription of rational antibiotic use, and antimicrobial stewardship Zhao et al. (2019) [20]. Though this study has added knowledge about local isolates of bacteria along with their susceptibility pattern, yet the retrospective design of this single center-based study is a limitation of this study. There is no information available about the clinical outcomes and molecular resistance mechanisms. Future studies may be carried out on a multicenter basis with prospective design and molecular resistance mechanism of the isolated strains may also be determined in future.

Conclusion

This retrospective study focuses on the microbiological characteristics and antimicrobial susceptibilities of respiratory infections at a tertiary care teaching hospital. Gram negative organisms, with *K. pneumoniae* being most prevalent, *P. aeruginosa* and *S. aureus* were other common bacteria found in respiratory infection cases. The highest antimicrobial efficacies were seen with Meropenem, Amikacin, and Piperacillin-Tazobactam, while significant resistance was found for commonly prescribed antibiotics like Ampicillin and Amoxicillin-Clavulanate. The presence of multidrug resistant organisms further indicates the increasing threat of antimicrobial resistance. This study clearly reflects that the importance of regular microbiological investigations and antimicrobial susceptibilities for appropriate treatment of respiratory infections is highly necessary.

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