Antibiogram Profile of Respiratory Pathogens and Identifying Predisposing Factors of Respiratory Tract Infections among Patients Visiting Bhaktapur Hospital

Shreemila Bajracharya[1†], Jharana Thapa[1†], Pushpa Thapa Magar[1†], Ananda Kumar Mandal[2], Niraj Manandhar[2], Avinash Chaudhary[1], Dinesh Dhakal[1], Upendra Thapa Shrestha[3*]
[1]Department of Microbiology, Sainik Awasiya Mahavidhyalaya, Bhaktpur, Nepal
[2]Bhaktapur Hospital, Bhaktapur, Nepal
[3]Central Department of Microbiology, Tribhuvan University, Kathmandu, Nepal
†These authors contributed equally.

*__Corresponding author:__ Upendra Thapa Shrestha, Central Department of Microbiology, Tribhuvan University, Kathmandu, Nepal, E-mail: upendrats@gmail.com

ABSTRACT

Objectives: To address the current problem of MDR infections and respiratory bacterial pathogens and identify the risk factors associated with respiratory tract infections (RTIs).

Methods: A total of 327 RTI specimens were cultured using a conventional microbiological method to identify potential bacterial pathogens. The isolates were then subjected to antibiotic susceptibility testing using modified Kirby Bauer's disc diffusion method. In addition, the risk factors associated with RTIs were obtained by direct interviews with patients using a structured questionnaire.

Results: The overall prevalence of RTIs among the study population was found to be 11.9%. The highest rate of infections was observed among patients of age group 61-70 years in both sexes and from ICU wards. Among the bacterial pathogens, *Acinetobacter baumannii* was isolated in the highest number followed by *Pseudomonas aeruginosa.* 64.2% *A. baumannii* isolates were resistant to Azithromycin but 100% sensitive to meropenem. *P. aeruginosa* resistance rate was 50% to Piperacillin-tazobactam, ciprofloxacin, Polymyxin B and Ceftriaxone, and had the highest MDR. Half of the *Staphylococcus aureus* isolates were MRSA. Risk factors such as previous infections, family history with RTIs, chronic use of antibiotics, and smoking were found to be significantly associated with RTIs.

Conclusion: We observed high MDR among the respiratory pathogens with growing resistance to β-lactam, macrolides, and Polymyxins whch necessitates alternative drugs in management. There is also need for targeted preventive strategies, prohibition of irrational use of antibiotics, and avoidance of exposure to risk factors such as smoking, outdoor air pollution, and chronic use of antibiotics.

Keywords: Respiratory tract infections, MDR, risk factors, Nepal

INTRODUCTION

Respiratory tract infections (RTIs) are major source of various health issues in humans. These infections can affect any part of the respiratory system. As the respiratory system is continually exposed to airborne germs and directly interacts with the environment, it is particularly susceptible to bacterial infections (Dewasy et al. 2021). Generally, infections of the respiratory tract are referred to as RTIs, while specific infections are distinguished as upper RTIs (URTIs) and lower RTIs (LRTIs) (Atray and Sheethal 2023).

Date of Submission: September 24, 2024
Date of Acceptance: December 15, 2024

DOI: https://doi.org/10.3126/tujm.v11i1.85475

URTIs are marked by temporary irritation and swelling of the upper airways, accompanied by a cough that does not indicate pneumonia, nor is it linked to any underlying medical condition that could lead to pneumonia. These infections impact areas such as the pharynx, larynx, sinuses, and nose (Morrison and Thomas 2023). URTIs primarily encompass tonsillitis, pharyngitis, rhinitis, laryngitis, sinusitis, and otitis media (Paudel et al. 2022). LRTI refers to inflammation of the respiratory tract, which encompasses pneumonia, lung abscess, bronchitis, bronchiectasis, and bronchiolitis (Khadka et al. 2022). Among these infections, LRTIs are major contributors to both illness and death, causing approximately 7 million deaths globally each year (Atray and Sheethal, 2023).

Common bacterial agents responsible for respiratory infections include *Klebsiella pneumoniae, Staphylococcus aureus, Streptococcus pneumoniae, Pseudomonas aeruginosa, Acinetobacter spp.,* and *Haemophilus influenzae.* The causes and clinical characteristics of respiratory diseases can differ based on factors such as age, gender, season, the specific at-risk population, and other variables (Chowdhury et al. 2023). Respiratory infections are a diverse and intricate set of diseases caused by various infectious agents, including viruses, bacteria, fungi, and parasites. Viral agents are responsible for a significant proportion of lower respiratory tract infections (LRTIs). The difficulty in distinguishing between different types of infections often leads to the unnecessary prescription of antibiotics, which can contribute to the development of drug resistance (Bhatta et al. 2023).

Multidrug-resistant bacteria are increasingly prevalent on a global scale as a result of the excessive and inappropriate utilization of antibiotics. The increase in multidrug-resistant bacteria represents a significant public health issue that compromises the effectiveness of antibiotics, prolongs the recovery process, and may lead to chronic colonization (Mbaya 2022). Therefore, the objective of this research was to analyze the antimicrobial susceptibility testing (AST) profile of bacterial pathogens derived from the respiratory tract and to identify the specific bacterial strains responsible for the infection. This investigation was carried out to acquire further insights into the prudent administration of antibiotics within healthcare settings, assess the various risk factors associated with respiratory infections, and determine their prevalence rates. Such insights are crucial for enhancing the accuracy of infection diagnosis and treatment.

METHODS

Study design, site, and population

A hospital-based cross-sectional study was conducted among the patients visiting Bhaktapur Hospital in the period between February to May. The clinical samples from the patients suspected of RTI were processed in the laboratory of Bhaktapur Hospital.

Inclusion and exclusion criteria

Both outpatients and inpatients (ICU, HDU, and medical ward) were included in this study. The patient's history with complaints of shortness of breath, cough with sputum, wheezing, fatigue, sore throat, and common cold were included. Those clinical samples that showed incomplete culture form, and contaminated specimens, without proper labeling including date, time, age, lab number, and gender were excluded. The sputum sample containing blood and saliva was excluded.

Sample types and size

The sputum, endo-tracheal swabs, and oral-pharyngeal swab samples were collected. The total sample size was 327 which was calculated according to the incidence of LRTI in developing countries that varied from 20–30%. Bhatta *et al.* (2023) Then, the final data was analyzed by using SPSS.

Specimen collection

Samples were collected by a well-trained person from the hospital. A clean dry wide-necked leakproof container was given to the patients, and they were requested to cough deeply to produce a sputum specimen without saliva soon after the patient woke up. The container was labeled appropriately with the patient's identification number, age, and gender. When instantaneous processing was not achievable, the samples were frozen at 4-6℃. For swabs, in a good light and using the handle of a spoon to depress the tongue, the inside of the mouth was examined. The healthcare professional looked for inflammation and the presence of any membrane, exudate, or pus. The affected area was swabbed using a sterile cotton wool swab, ensuring not to contaminate the swab with saliva, and it was returned to its sterile container and delivered to the laboratory within two hours of collection (Cheesbrough 2006).

Culture of specimen

Sputum samples were washed in sterile saline. Then, with the help of an inoculating loop, sputum was inoculated on blood, chocolate, and MacConkey agar plates and incubated at 37°C for 24 hrs. Chocolate agar plates were incubated in an atmosphere enriched with CO_2. Swabs were cultured on blood agar and Mac Conkey agar at 37°C for 24 hours. The grown colonies were also subcultured on nutrient agar plates (Cheesbrough 2006).

Identification of the isolates

The identification of significant bacterial isolates was done by using microbiological techniques as described in Monica Cheesbrough which entailed the morphological appearance of the colonies, Gram-staining reactions, and biochemical properties.

Antibiotic susceptibility test

Antibiotic susceptibility test for isolated organisms was done on Muller Hinton Agar (MHA) plates by using the Modified Kirby Bauer Disc Diffusion technique following the Clinical and Laboratory Standard Institute (CLSI) guidelines (CLSI, 2020). Interpretation as 'Sensitive' or 'Resistant' was completed based on the diameters of zones of inhibition of bacterial growth Cheesbrough, (2006). Also, the isolates resistant to three or more antimicrobial classes were considered multidrug-resistant (Magiorakos et al. 2012).

Ethical consideration

The study was approved by the Institutional Review Committee (IRC), Institute of Science and Technology, Tribhuvan University (IRC/IoST-24-0020). Written consent was taken from all the participants before the collection of samples and the data.

RESULTS

Bacterial growth in different respiratory samples

Among the total 327 samples, bacterial pathogens were detected in 11.9% of the cases (Figure 1).

Age and gender-wise distribution of bacterial growth

Among different age groups, the highest bacterial growth was observed among the 61-70 age group for both males (28.5%) and females (27.7%) followed by the 71-80 age group for both males (19%) and females (22.2%). Similarly, males had a higher infection rate (14%) compared to females (10.16%). However, the result was not statistically significant ($p > 0.05$) (Table 1).

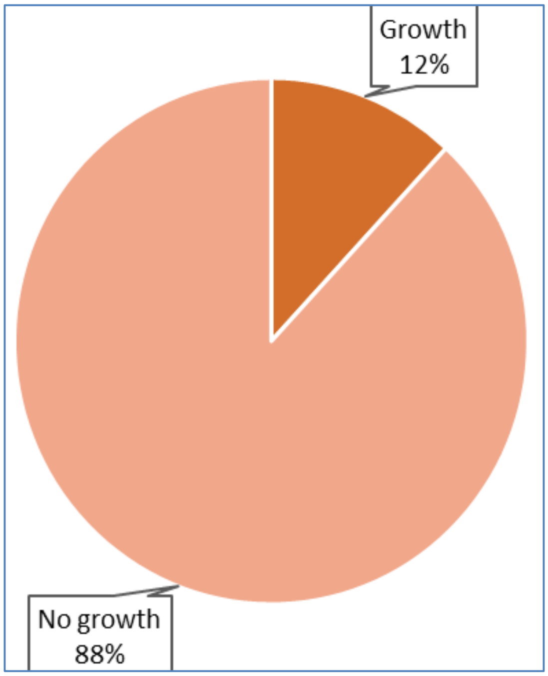

Figure 1: Bacterial growth in respiratory samples

Ward-wise distribution of bacterial growth

Samples from the ICU ward showed the highest rate of bacterial growth (17.6%) followed by medical ward (12.8%) and then the HDU ward (2.7%) (Figure 2).

Sample-wise distribution of bacterial growth

Endotracheal (ET) swabs exhibited the highest bacterial growth (66.7%) followed by sputum (MP) samples (17.1%). Throat swabs and sputum (MS) samples showed no bacterial growth. The results were not statistically significant ($p > 0.05$) (Table 2).

Distribution of bacterial growth based on cell count and KOH mount

Samples with epithelial cell numbers >10 and pus cell >25 showed the highest positive results (21.7%), followed by EP< 10, Pus> 25 (18.5%) ($p > 0.05$). However, the KOH mount showed that samples without fungal elements had higher positive results (10%) compared to those with fungal elements (7%), giving a statistically significant finding ($p < 0.05$) (Table 3).

Frequency of Gram-positive and Gram-negative bacteria

Among the 39 isolates, Gram-negative bacteria were predominant (84.60%) to Gram-positive bacteria (15.40%).

Distribution of bacterial isolates

The most predominant bacteria among the 39 bacterial isolates were *Acinetobacter baumannii* (35.9%), followed by *Pseudomonas aeruginosa (*30.77%), *Staphylococcus aureus (*15.38%), *Klebsiella pneumoniae (*12.83%) and *Klebsiella oxytoca* (5.13%) (Figure 3).

Antibiotic resistance pattern of bacterial isolates

Among the isolates*, A. baumannii* were highly resistant to Azithromycin (64.2%) followed by Polymyxin B (57.12%), ciprofloxacin (42.84%) whereas least resistant to Amoxicillin-clavulanate (7.14%). *P. aeruginosa* were highly resistant to azithromycin (50%) and ceftriaxone (50%) while least resistant to meropenem (8.33%), Piperacillin-tazobactam (8.33%). Similarly, *K. pneumonia* was highly resistant to azithromycin (60%) followed by linezolid (40%). *K. oxytoca* was resistant to ciprofloxacin (50%), amikacin (50%), and polymyxin B (50%). For *S. aureus,* they were highly resistant to ciprofloxacin (83.33%) followed by Cloxacillin (66.67%) in which out of 6 strains, 3(50%) were MRSA (Table 4).

Rate of Methicillin-Resistant *Staphylococcus aureus* (MRSA)

The rate of both MRSA and MSSA was found to be 50% among the six strains of *S. aureus*.

Multidrug-resistant (MDR) strains

Among the different bacterial isolates, the highest MDR strains were *P. aeruginosa (*66.67%), followed by *K. pneumoniae* (60%), *A. baumanni* (57.14%) and *K. oxytoca* (50%) (Table 5).

Predisposing factors affecting RTIs

Out of 327 patients, 17.3% RTIs were found among the patients with a family history of chronic RTIs ($p < 0.05$). Past infections and regular antibiotic users were found to be the significant predisposing factors for RTIs which account for 19.8% and 27.0% respectively ($p < 0.05$). Patients with symptoms such as shortness of breath, chest tightness, and cough had an infection rate higher (12.4%) as compared to those without any symptoms ($p > 0.05$). Exposure of patients to occupational hazards resulted in a slightly higher rate of infections (12.3%) while indoor pollution exposure had a lower infection rate (11.6%) as compared with the exposed population. Patients with a smoking habit were more susceptible to infection (16.5%) compared to those without a smoking habit (9.6%) ($p < 0.05$). Similarly, non-former smokers had a high infection rate (12.4%). Additionally, passive smokers were found to be more infected (12.8%) compared to non-passive smokers (10.7%) ($P > 0.05$) (Table 6).

Table 1: Age and gender-wise distribution of bacterial growth

Gender / Age group of patients	Male*		Female**	
	No growth (%)	Growth (%)	No growth (%)	Growth (%)
NB* - 10	1 (0.7)	0	0	0
11 - 20	2 (1.5)	1 (4.7)	1 (0.6)	1 (5.5)
21 - 30	6 (4.6)	4 (19.0)	6 (3.7)	1 (5.5)
31 - 40	11 (8.5)	2 (9.5)	10 (6.2)	0
41 - 50	14 (10.8)	2 (9.5)	11 (6.9)	3 (16.6)
51 - 60	20 (15.5)	2 (9.5)	38 (23.8)	4 (22.2)
61 - 70	28 (21.7)	6 (28.5)	43 (27.0)	5 (27.7)
71 - 80	36 (27.9)	4 (19.0)	42 (26.4)	4 (22.2)
81 - 90	9 (6.9)	0	8 (5.0)	0
100	2 (1.55)	0	0	0
Total	**129 (86)**	**21 (14)**	**159 (89.83)**	**18 (10.16)**

NB = new born; *growth vs. gender (p = 0.287); **growth vs. age (p = 0.145)

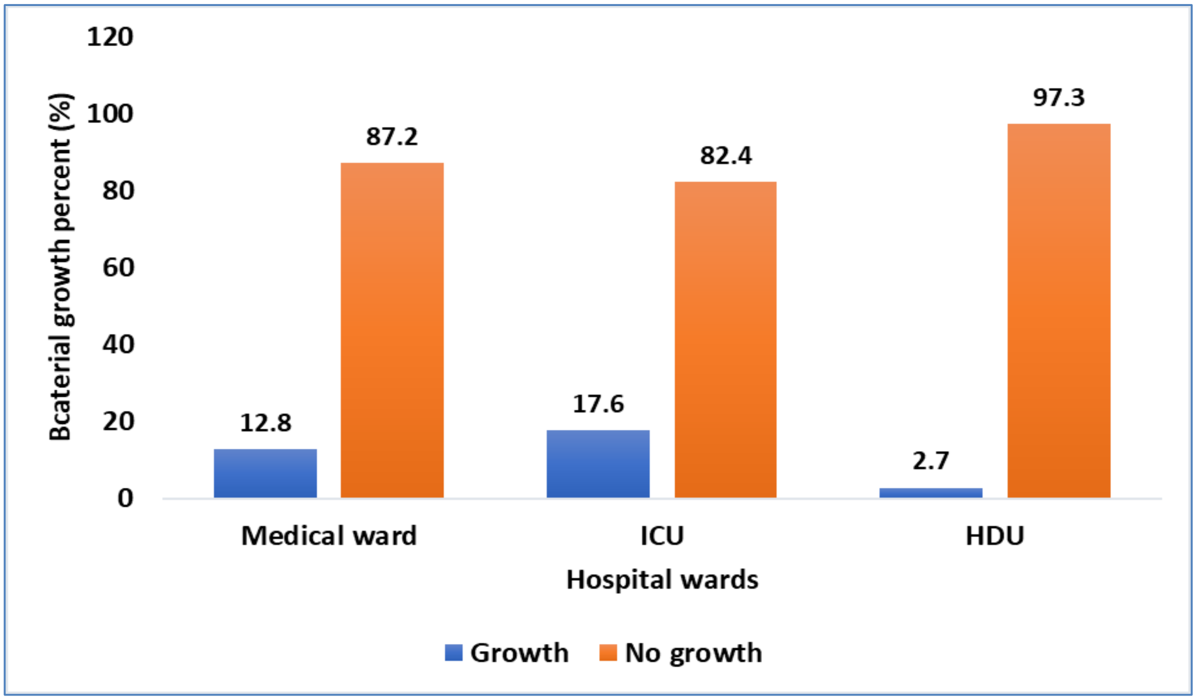

Figure 2: Ward-wise distribution of bacterial growth

Table 2: Sample-wise growth pattern of bacterial pathogens

Types of samples	No growth (%)	Growth (%)	p-value
Sputum (MP)	68 (82.9)	14 (17.1)	0.054
Sputum (M)	114 (91.2)	11(8.8)	
Sputum (P)	94 (88.7)	12 (11.3)	
Sputum (MS)	9 (100.0)	0	
ET swab	1 (33.3)	2 (66.7)	
Throat swab	2 (100.0)	0	
Total	**288 (88.1)**	**39 (11.9)**	

Table 3: Cell count/ KOH mount of RTI-infected patients

Parameters		**Sputum Culture**		**p-value**
		No growth (%)	**Growth (%)**	
No. of epithelial and pus cells	EP < 10, Pus >25	44 (81.5)	10 (18.5)	0.162
	EP < 10, Pus <25	71 (92.2)	6 (7.8)	
	EP > 10, Pus <25	135 (90.6)	14(9.4)	
	EP > 10, Pus >25	18 (78.3)	5 (21.7)	
KOH mount	Fungal elements seen	119 (93)	9 (7)	0.023
	Fungal elements not seen	70 (89.7)	8 (10.3)	

Table 4: Antibiotic resistance pattern of bacterial isolates, N (%)

Antibiotics category	Antibiotics	*A. baumannii*	*P. aeruginosa*	*K. pneumonia*	*K. oxytoca*	*S. aureus*
Macrolides	Azithromycin	9 (64.2)	6 (50)	3 (60)	0	3 (50)
Penicillin + β - lactamase inhibitors	Amoxicillin-clavulanate	1(7.14)	4 (33.33)	0	0	3 (50)
	Cloxacillin	0	1(8.33)	1 (20)	0	4 (66.67)
Antipseudomonal penicillin + β - lactamase inhibitors	Piperacillin-tazobactam	0	1(8.33)	0	0	0
Aminoglycosides	Amikacin	3 (21.42)	3 (25)	2 (40)	1 (50)	0
	Gentamycin	0	0	0	0	1 (16.67)
Fluoroquinolones	Ciprofloxacin	6 (42.84)	2 (16.67)	0	1 (50)	5 (83.33)
	Levofloxacin	4(28.57)	2 (16.67)	0	0	2 (33.33)
Linosamides	Clindamycin	0	4 (33.33)	1 (20)	0	1 (16.67)
Cephalosporins	Cefoperazone-Sulbactam	0	1(8.33)	0	0	0
	Ceftazidime	4 (28.57)	1(8.33)	1 (20)	1 (50)	0
	Ceftriaxone	3 (21.42)	6 (50)	1 (20)	0	2 (33.33)
Glycylcylines	Tigecycline	0	2 (16.67)	1 (20)	0	0
Oxazolidinones	Linezolid	2 (14.29)	6 (50)	2 (40)	0	1 (16.67)
Polymyxin	Polymyxin B	8 (57.12)	6 (50)	2 (40)	1 (50)	2 (33.33)
Carbapenem	Meropenem	0	1(8.33)	1 (20)	1 (50)	0
Antistaphylococcal β lactam	Cefoxitin	0	0	0	0	3 (50)

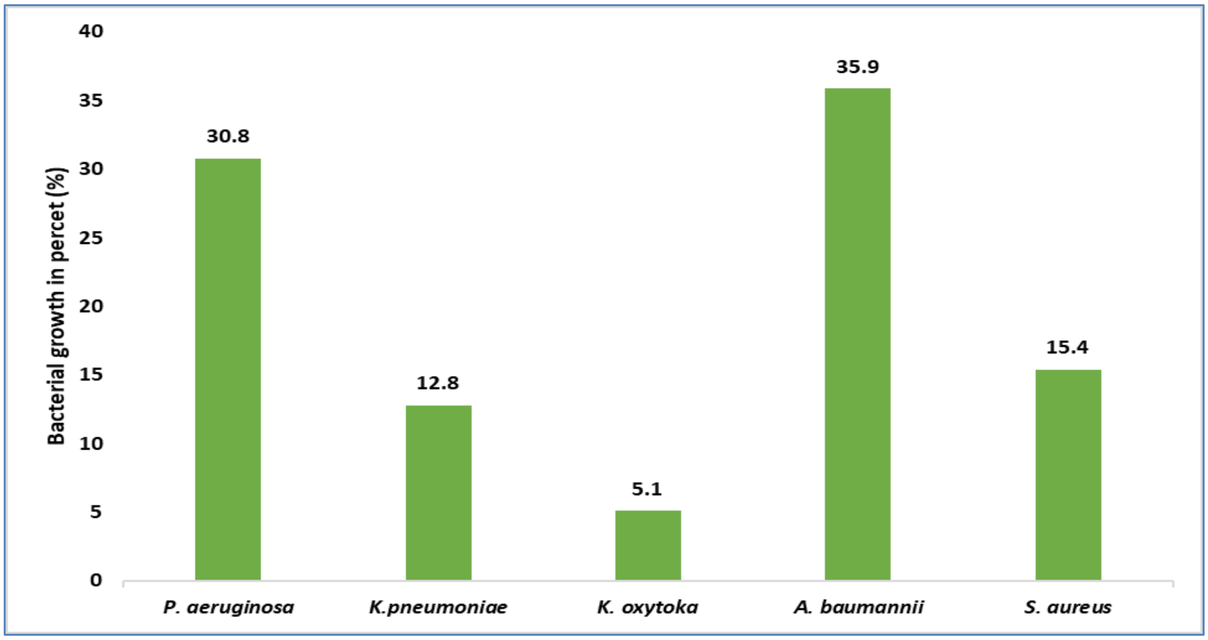

Figure 3: Growth percentage of isolated bacteria

Table 5: Number of MDR strains among different bacterial pathogens

Organisms	Total Number	MDR Number (n)	Percentage(%)
P. aeruginosa	12	8	66.67
A. baumannii	14	8	57.14
K. pneumoniae	5	3	60
K. oxytoca	2	1	50
S. aureus	6	3	50
Total	**39**	**23**	**58.97**

Table 5: Risk factors of RTIs

Parameters	Frequency	Positive [n(%)]	Negative [n(%)]
Family history of RTI			
• With a family history	98	17 (17.3)	81 (82.7)
• Without family history	229	22 (9.6)	207 (90.4)
Past infections (pneumonia, bronchitis, sinusitis)			
• With past infections	81	16 (19.8)	65 (80.2)
• With no past infections	246	23 (9.3)	223 (90.7)
Long-term use of antibiotics			
• Antibiotic users	89	24 (27.0)	65 (73.0)
• Antibiotic non-users	238	15 (6.3)	223 (93.7)
Symptoms (shortness of breath, chest tightness, cough)			
• with symptoms	283	35 (12.4)	248 (87.6)
• without symptoms	44	4 (9.1)	40 (90.9)
Occupational hazards			
• Patients being exposed	122	15 (12.3)	107 (87.7)
• Not exposed	205	24 (11.7)	181 (88.3)
Indoor pollution			
• Exposed	95	11 (11.6)	84 (88.4)
• Not exposed	232	28 (12.1)	204 (87.9)
Smoking habits			
• Smokers	109	18(16.5)	91(83.5)
• Non-smokers	218	21(9.6)	197(90.4)
• Former smokers	61	6 (9.8)	55 (90.2)
• Non-former smokers	266	33 (12.4)	233 (87.6)
• Passive smokers	196	25 (12.8)	171 (87.6)

DISCUSSION

In this study, 39 samples (sputum and ET swabs) yielded significant bacterial growth (11.9%) out of 327 samples. A similar study conducted by Subedi et al.(2019) reported higher bacterial growth (30.2%). Prior research reported 24.62% bacterial growth. The culture positivity of the samples may be influenced by different factors such as type of specimen, time of specimen transport, and number of organisms present in the sample. One of the significant causes of negative cultures in RTI infection may be the previous use of antibiotics (Nepal et al. 2018).

The study revealed a significant correlation between age groups and the rate of bacterial infections. The highest rate of infection was observed in the age group of 61-70 years followed by the 71-80 age group. Previous studies showed infection rates higher in the 60-69 (19.7%) and 70-79 (19.7%) age groups (Dewasy et al. 2021) indicating increased susceptibility to infections in older adults reported 46-55 age group to be highly affected with infection rate of 34.1% (Vaish 2021). Another study conducted by Kalgo et al. showed increased prevalence in the 20-39 (43.5%) age group which was very different from our findings (Kalgo et al. 2022). The higher infections among the older population might be due to a decline in immune functioning and exposure to chronic infections like diabetes, and hypertension increasing susceptibility to infections.

In our study, males showed a higher infection rate (14%) than females (10.16%). A similar study carried out in Eastern Nepal also reported a high prevalence in males (55.64%) compared to females (44.35%) Dewasy et al.(2021). Likewise, the study by Nurahmed et al.(2020) also reported higher RTIs infections in males (44.6%) than in females (21.1%). However, a study by Mbaya A, (2022)reported females were more highly infected (53.1%) than males (46.9%) giving different results. The increased infection rate among the male population may be associated with exposure to various risk factors such as cigarette smoking or cultural differences within various communities as well as frequent exposure to environmental pollutants (Nurahmed et al. 2020).

According to our study, the highest rate of infection was observed among the patients from the ICU ward (17.6%), followed by the medical ward (12.8%) and the HDU ward (2.7%). In a previous study, patients from the medical ward had the highest infection rate (76.1%) followed by the ICU ward (7.1%) (Dewasy et al. 2021). The elevated bacterial growth rate in the ICU ward corresponded with the well-established risks of hospital-acquired infections in critical care settings. This highlights the necessity for strict infection control protocols and ongoing staff training in these environments.

Endotracheal swabs showed the highest bacterial growth (66.7%) followed by muco-purulent sputum samples (17.1%). In a study carried out by Subedi et al. the frequency of pathogens was greater in the sputum sample (29.13%) as compared to throat swabs (1.07%) (Sunedi et al. 2019). Endotracheal swabs demonstrated the highest level of bacterial growth, indicating that invasive respiratory procedures might increase the risk of bacterial infections. This underscores the importance of aseptic techniques and careful monitoring of patients undergoing these procedures.

Out of 303 sputum samples for which cell count was performed, samples with epithelial cells more than 10 and pus cells more than 25 gave the highest positive results (21.7%) followed by the samples with epithelial cells less than 10 and pus cells more than 25 (18.5%). Among the 206 sputum samples for which KOH mount was performed, 128 samples showed fungal elements, with 7% showing bacterial growth. Conversely, of the 78 samples without fungal elements, 10% still yielded positive results. This suggests that bacterial infections are more likely in samples with specific cell counts and the absence of fungal elements, pointing towards the complex interplay between bacterial and fungal pathogens in respiratory infections.

Gram-negative bacteria were more predominant (84.60%) than Gram-positive bacteria (15.40%). A prior study showed a higher prevalence of Gram-negative bacteria as compared to Gram-positive bacteria Subedi et al. (2019). A higher incidence of Gram-negative bacteria (80.9%) was reported by Bhatta et al. (2023) in contrast to Gram-positive bacteria (19.1%) which was similar to our findings. A study by Vaish reported a higher prevalence of Gram-negative bacteria (74.1%) than Gram-positive bacteria (25.8%) (Vaish 2021). High occurrence of Gram-negative bacteria may be because they can be commonly found in hospital environments and their ability to thrive in these environments can contribute to their high prevalence in clinical infections.

Among the Gram-negative bacteria, the most predominant isolate was *Acinetobacter baumannii (35.9%),* followed by *Pseudomonas aeruginosa (*30.77%) whereas *Klebsiella oxytoca* (5.13%) was the least occurring. In research conducted by Bhatta et al., Gram-negative bacteria were predominantly associated with respiratory tract infections with *Acinetobacter* species (29.4%) as the most common pathogen (Bhatta et al. 2023). However, the study carried out by Kalgo et al.(2022) showed *S. aureus* (31.1%) as the predominant one. The predominance of gram-negative bacteria, particularly *Acinetobacter baumannii* and *Pseudomonas aeruginosa*, is concerning due to their known resistance to multiple antibiotics.

P. aeruginosa was highly resistant to Polymyxin B (50%)

followed by Ceftriaxone (50%) and Linezolid (50%). Similar findings were reported by Subedi et al. (2019) in which the higher sensitivity of *P. aeruginosa* was observed to Ciprofloxacin (71%) followed by Piperacillin-tazobactam (42%). However, Sherchan and Humagain, (2021) had a different result which reported that 5% were resistant to Piperacillin-tazobactam, Amikacin, and Carbapenem Sherchan and Humagain, (2021). In recent research by Hoang et al. (2024), *P. aeruginosa* had high resistance to fluoroquinolones (72.1%) and carbapenem (66.7%). Likewise, *A. baumannii* was highly resistant to Azithromycin (45%) whereas sensitive to Meropenem (70%) followed by Piperacillin-tazobactam (65%) which is very different than the study by Sherchan and Humagain, (2021) which showed 88.89% of *A. baumannii* are resistance to Piperacillin-tazobactam and Cefoperazone-sulbactam recent study by Le et al. (2024) *A. baumannii* were high resistant to Cephalosporins (90%) and Fluoroquinolones (80%) and least resistance to Colistin (12.5%) and Amikacin (29.6% Le et al. (2024).

In this study, *K. pneumoniae* were highly sensitive to Amoxicillin-clavulanate (50%) and Ciprofloxacin (50%) while least resistance to Colistin (30%) and Azithromycin (30%) which was similar to findings by Subedi et al. (2019) that showed sensitivity to Ciprofloxacin (43.5%). The study by Sherchan and Humagain, (2021)showed different results than our study where *K. pneumoniae* were resistant to Amoxicillin-clavulanate (84.34%) and Ciprofloxacin (80.72%). However, in a recent study by Le et al. (2024). *K. pneumoniae* were highly resistant to penicillin class mainly Ampicillin (100%), and least resistant to Colistin (11.6%) and Polymyxin (7.1%). Likewise, *K. oxytoca* was sensitive to Piperacillin-tazobactam (20%) while resistant to Amikacin (90%), Ciprofloxacin (90%), and Meropenem (90%) which is higher than the study by Sherchan and Humagain, (2021) showing resistance to Ciprofloxacin (83.33%) and Piperacillin-tazobactam (83.33%). *S. aureus* were highly sensitive to Gentamycin (50%) and Amikacin (50%) followed by Clindamycin (40%) which is comparable to findings by Subedi et al. (2019)with Clindamycin (50%) and Erythromycin (50%). Out of six strains of *S. aureus,* 50% were MRSA which is comparable to the study by Bhatta et al. (2023) where 54.5 % were MRSA.

In this study, the number of MDR strains was most predominant among *P. aeruginosa* (66.67%) followed by *K. pneumoniae* (60%), *A. baumannii* (57.14%), *K. oxytoca* and *S. aureus* (50%). which is higher than the study carried out by Nurahmed et al. (2020), Nepal et al. (2018), and Bhatta et al. (2023) had MDR isolates(40%). The increasing rate of MDR among bacterial pathogens from RTIs might be due to the irrational use of antibiotics, misuse of antibiotics in empirical therapy, and overuse of antibiotics in viral and other non-bacterial infections.

Out of 327 patients, 17.3% of those with a family history of chronic RTIs experienced severe infections. Additionally, patients who had been previously infected with RTIs showed a higher incidence of infections. Similarly, patients who used long-term antibiotics were prone to respiratory infections. These factors should be carefully considered in clinical practice to identify high-risk patients and implement preventive measures. The patients experiencing symptoms such as shortness of breath, chest tightness, and cough were more susceptible to respiratory infections. A prior study conducted by Mbaya et al. (2022) reported cough as the sole factor accounting for 52.6 % of the total cases followed by common cold Mbaya *et al.* (2022). 12.3% of those who were exposed to occupational hazards (like chemicals, dust, fumes, etc.) were found to be highly infected. This is lower than the infection rate reported in a study by Nurahmed et al. (2020), which was 76.2%. On the other hand, 11.6% of patients exposed to indoor pollution (such as smoke from cooking, heating, or poor ventilation) were found to be infected. In a study carried out by Nurahmed et al. (2020), the majority of female participants were infected and were exposed to indoor pollution.

In this study, patients with smoking habits consuming more than five pieces of cigarettes per day showed a higher rate of infection (16.5%) compared to that of non-smokers (9.6%). Nurahmed et al. (2020) reported smokers to be highly infected by 58.3% and non-smokers by 27.5% which closely aligns with our study. Likewise, 16.5% of the patients consuming tobacco were highly infected with RTI in comparison to non-tobacco consumers (10.2%). This highlights the role of smoking as a significant risk factor for respiratory infections and reinforces the need for smoking termination programs to reduce respiratory infection rates.

Conclusion

The study found that RTIs are more common in men than in women and that 58.9% of the 39 bacterial isolates were multi-drug resistant (MDR). *P. aeruginosa*, *A. baumannii, K. pneumoniae,* and *K. oxytoca* showed high resistance to macrolides and polymyxins, while *S. aureus* was mainly resistant to penicillin and β-lactamase inhibitors. Consequently, β-lactams, macrolides, and polymyxins are not recommended for treating RTIs. Effective management of RTIs requires thorough microbiological diagnosis and susceptibility testing, along with ethical antibiotic use to reduce resistance. Risk factors for RTIs include a family history of RTIs, daily cigarette smoking, and past infections. Addressing these factors through early detection and control can help reduce RTI prevalence.

ACKNOWLEDGEMENTS

We express our sincere thanks to the laboratory staff of Bhaktapur Hospital and the participants for providing respiratory samples and the required information.

CONFLICT OF INTEREST

The authors declared no conflict of interest.

REFERENCES

Atray D, Sheethal S. (2023). Bacteriological profile and antibiotic susceptibility patterns of Gram-negative bacilli isolated from lower respiratory tract infections. *Indian Journal of Medical Specialities,* **14**(1), 31.

Bhatta D R, Hamal D, Shrestha R and Nayak N. (2023). Antibiotic Resistance Patterns of Bacterial Pathogens Associated with Lower Respiratory Tract Infections. *Nepal Journal of Medical Sciences,* **8**(1), 5–11.

Cheesbrough M. (2006). District Laboratory Practice in Tropical Countries (2nd ed). *Cambridge University Press.*

Chowdhury M, Jobayer M and Shamsuzzaman S. Bacteriological Profile and Antibiogram of Respiratory Tract Infections in a Tertiary Care Hospital, Bangladesh. *Bangladesh Medical Res Counc Bull 2023;* ***49****(1): 15-21*

CLSI. Clinical and Laboratory Standard Institute: Performance standards for antimicrobial susceptibility testing .30th Edition ed. Wayne, PA 19087 USA M 100.: Clinical and Laboratory Standard Institute antimicrobial susceptibility testing standards M02, M07 and M11;2020.

Dewasy B, Singh TSK, Singh R and KafleT (2021). Prevalence of Typical Bacterial Load and their Susceptibility to Antimicrobial Agents in Patients with Lower Respiratory Tract Infections in Eastern Nepal. *International Journal of Innovative Science and Research Technology* ***6(****12).*

Kalgo Z M, Amin B M and Mohammed B. (2022). Antimicrobial Susceptibility Pattern of Bacterial Pathogens Isolated from Patients with Lower Respiratory Tract Infection in Kebbi State, Nigeria. *JMIR Publications 2022*

Khadka S, Barakoti A, Adhikari R P, Khanal L K and Sapkota J. (2022). Positive Bacterial Culture among Lower Respiratory Tract Specimens of Patients in a Tertiary Care Centre: A Descriptive Cross-sectional Study. *Journal of Nepal Medical Association*, **60(**248), 384–388.

Le H H, Nguyen A V, Vu L H, Nguyen V T H, Pham H Q, Le H V, Nguyen S T, Le H T, Dinh H V, Le N V, Le T D, Le M N, Nguyen V H, Hoang K T and Le H H L. (2024). Antimicrobial Resistance Patterns of Common Gram-Negative Microorganisms Isolated from Patients with Lower Respiratory Tract Infections in a Teaching Hospital in Vietnam. *Japanese Journal of Infectious Diseases*, **77(**3), 144–154.

Magiorakos A P, Srinivasan A, Carey R B, Carmeli Y, Falagas M E, Giske C G, Harbarth S, Hindler J F, Kahlmeter G, Olsson-Liljequist B, Paterson D L, Rice L B, Stelling J, Struelens M J, Vatopoulos A, Weber J T and Monnet D L (2012). Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: An international expert proposal for interim standard definitions for acquired resistance. *Clinical Microbiology and Infection*, **18(**3), 268–281.

Mbaya A H. (2022). Antimicrobial Drug Prescription Patterns for Respiratory Tract Infections in Military Hospitals Lagos Nigeria. *The Nigerian Journal of Pharmacy*, **56(**2).

Nepal R, Shrestha B, Joshi D M, Joshi R D, Shrestha S and Singh A. (2018). Antibiotic Susceptibility Pattern of Gram-negative Isolates of Lower Respiratory Tract Infection. *Journal of Nepal Health Research Council*, **16(**1), 22–26.

Nurahmed N, Kedir S, Fantahun S, Getahun M, Mohammed A, Mohammed F, Arega T and Desta K. (2020). Bacterial profile and antimicrobial susceptibility patterns of lower respiratory tract infection among patients attending selected health centers of Addis Ababa, Ethiopia. *The Egyptian Journal of Chest Diseases and Tuberculosis*, **69(**2), 399.

Paudel G, Amatya N and Adhikari B. Nasal colonization by potential bacterial pathogens in healthy kindergarten children of Nepal: A prevalence study. *National library of medicine* **12***(1):86-98.*

Sherchan J B and Humagain S. (2021). Antimicrobial Susceptibility Pattern of Gram-Negative Bacteria Causing Lower Respiratory Tract Infections in Kathmandu University Hospital. *Journal of Nepal Health Research Council*, **18(**4), 661–666.

Subedi D, Devkota S P, Bhatta D R, Sharma B K, Paudel A, Gurung K and Gautam D. (2019). Antibiotic Susceptibility Profile of respiratory pathogens obtained at tertiary care hospital from western Nepal. *Nepal Journal of Biotechnology,* **7**(1), 1–7.

Vaish R. (2021). Antibiotic susceptibility of bacterial strains and bacteriological profile from patients with lower respiratory tract infections in a teaching hospital. *Panacea Journal of Medical Sciences*, **11(**2), 326–330.