



ORIGINAL RESEARCH PAPER

Clinical Microbiology

BACTERIAL ISOLATES OF THE RESPIRATORY TRACT INFECTION AND THEIR CURRENT SENSITIVITY PATTERN AMONG PATIENTS ADMITTED IN TERTIARY CARE HOSPITAL.

KEY WORDS: Isolate, Bacteria, Respiratory tract infections, antibiotic susceptibility pattern.

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ABSTRACT

Background & Objectives: This study was aimed to observe the susceptibility pattern of bacterial isolates from respiratory tract infection (RTI). Respiratory tract infection is considered as one of the major public health problems and a leading cause of morbidity and mortality in many developing countries. Respiratory tract is the part of the human system that plays a vital role in breathing processes. In human, the respiratory system can be subdivided into an Upper respiratory tract and a Lower respiratory tract based on anatomical features. The respiratory tract is constantly exposed to microbes due to the extensive surface area. **Methods:** The present study was conducted retrospectively for a period of one year November 2021 to October 2022. All respiratory specimens included Sputum, BAL, throat swab; endotracheal aspirate specimens were collected aseptically from patients and cultured on the appropriate bacteriological media (Blood agar, MacConkey agar & Chocolate Agar). Bacterial isolates were identified by biochemical tests and antimicrobial susceptibility performed by standard methods as per CLSI 2022. **Results:** 152 (72.3%) of total 210 samples were positive for bacterial culture. 126 (82.8%) were gram negative bacilli (GNB) and 26 (17.1%) were gram positive cocci (GPC). The predominant pathogen isolated was *K. pneumoniae* 46 (30.2%) followed by *Escherichia coli* 28 (18.4%). The overall susceptibility of GNB was highest towards Imipenem, Meropenem followed by Piperacillin tazobactam and Amikacin. Gram positive organisms exhibited highest susceptibility towards Vancomycin and Linezolid. **Conclusion:** Imipenem is the most sensitive antibiotic followed by Piperacillin tazobactam and Amikacin which can be used for empirical therapy for respiratory tract infections (RTI). The antibiotic therapy should be modified as per the culture and sensitivity report. Regular determinations of the type of bacterial pathogens and updation of antibiogram must be followed in every institution to aid in better patient management by helping the clinician in the judicious use of antibiotics.

INTRODUCTION

Respiratory tract infection (RTI) is considered as one of the major public health problems and a leading cause of morbidity and mortality in many developing countries (Bipin Prajapati *et al.*, 2019) [1]. Infection can arise from other people by cross-infection or even from an environmental source. The microorganisms larger than 10µm usually are trapped by the hair and cilia lining the nasal cavity. Coughing and sneezing reflexes clear the respiratory system of microorganisms by expelling them forcefully from the lungs through the mouth and nose respectively (Prescott *et al.*, 2005) [2]. The most infections are limited to the upper respiratory tract and only 5% involve the lower respiratory tract, respectively. Upper Respiratory Infections (URTIs) involve the nasal passages, pharynx, tonsils and epiglottis. The nasal discharge associated with colds contains virus particles, dead cells from the nasal mucosa and bacteria. Lower respiratory tract infections (LRTIs) involve the bronchi and alveoli. They include two serious conditions—acute bronchitis and pneumonia. Inflammation of the lung is a serious condition, responsible for most of the deaths caused by infection of the respiratory tract, especially in adults and infants (Manikandan C. 2013) [3]. Each type of RTI varies in the epidemiology, pathogenesis, clinical presentation, and outcome. The major respiratory pathogens are Gram negative bacilli (GNB) like *Klebsiella pneumoniae* (*K.pneumoniae*), *Escherichia coli* (*Esch.coli*), *Pseudomonas aeruginosa* (*P.aeruginosa*), *Acinetobacter* species and gram positive

organisms like *Streptococcus pneumoniae* (*Str.pneumoniae*), *Staphylococcus aureus* (*S.aureus*) [4]. However, results from various surveillance studies show wide variations in the prevalent etiological agents and their antimicrobial resistance patterns, both geographically and over time. The dramatic rise in the antimicrobial resistance among the respiratory pathogens, presumably due to the prophylactic administration of antibacterial therapy even before the availability of the culture reports, is a matter of potential concern worldwide. Failure to de-escalate the therapy after getting the culture and sensitivity report is another important reason for the drug resistance. In this context, management of these infections has become a challenge to the physicians. Hence the present study was conducted to investigate the bacterial etiology of LRTIs & URTIs in our institution and to update the clinicians on the current antibiotic susceptibility pattern of these pathogens. [5]

MATERIALS AND METHODS

Study Population:

In the present study, a total of 210 clinical samples were collected from patients who admitted in L.N. Medical College & J. K. Hospital Bhopal Madhya Pradesh, India. All patients had clinical evidence of respiratory tract infections, as determined by the treating doctors. Only a single positive culture per patient was included in the analysis.

Specimen Collection:

The specimens were collected aseptically from 210 (Sputum 86, Throat swab 30, BAL 57, ET Secretion 37 samples). All patients were instructed on how to collect the sputum samples aseptically and taken to the laboratory immediately for analysis. The sputum samples were collected into well-labeled sterile, universal container and using a sterile cotton swab, inner surface of the infected throat was swabbed gently and then the swabs were transported to the Microbiology laboratory. The study was carried out for one year between November 2021 to October 2022.

Bacteriology:

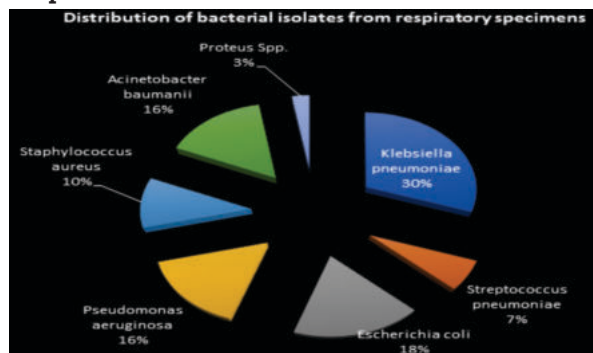
In the laboratory, each sample was inoculated on McConkey agar, Chocolate agar and Blood agar. The inoculum on the plate was streaked out for discrete colonies with a sterile wire loop. The culture plates were incubated at 37°C for 24 hours and observed for growth through the formation of colonies. All the bacteria were isolated and identified using morphological, microscopy and biochemical tests (Indole, Methyl red, Urease, Citrate, Mannitol motility, Triple sugar iron, Catalase, Coagulase and Oxidase test) following standard procedures described by (Sharma 2008)¹⁶

Antibiotic Susceptibility Testing:

The antimicrobial sensitivity of the test strains of twenty one (21) antibacterial drugs (HiMedia Laboratories Pvt Ltd) was done using the Kirby-Bauer disk diffusion Method. The commercial available antibiotic discs used for the study were Amikacin (30µg) Gentamicin (10µg) Imipenem (10µg) Meropenem (10µg) Piperacillin-Tazobactam (10µg) Ampicillin-Sulbactam (10µg) piperacillin (100µg) cefotaxime (30µg) ceftazidime (30µg) amoxicillin-clavulanate (10µg) ciprofloxacin (5µg) levofloxacin (5µg) Cotrimoxazole (1.2/23.75mcg) cefixime (5µg) cefepime (30µg) Penicillin (10 units) Cefoxitin (30µg) Erythromycin (15µg) Clindamycin (2µg) Tetracycline (30µg) Linezolid (30µg). The isolates were aseptically lawn cultured onto prepared Mueller Hinton agar. Then using sterile forcep, standard antibiotic discs were placed on the surface of the inoculated agar plates. Then incubate at 37°C for overnight incubation. After overnight incubation, the plates were observed for the zones of inhibition. Sensitivity of the isolates were determined by measuring the diameter of each zone of inhibition around each disc and the values obtained were compared with the CLSI guidelines 2022.

RESULT:

Out of 210 respiratory samples, 152 (72.3%) were positive for bacterial culture. Remaining samples yielded normal pharyngeal flora/ mixed flora/no growth/fungal growth. Among the bacterial isolates 126 (82.8%) were GNB and remaining 26 (17.1%) were gram positive cocci. The predominant pathogen isolated was Klebsiella pneumoniae (30.2%) followed by Escherichia coli (18.4%). Among gram positive bacteria, Staphylococcus aureus (10.5%) organisms followed Streptococcus pneumoniae (6.57%). The rate of isolation of different bacterial isolates is shown in **Table 1 & Graph 1**

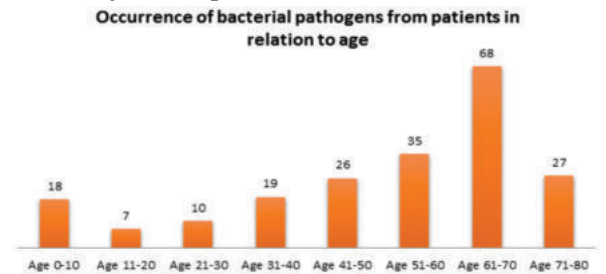


Graph 1: Distribution of bacterial isolates from respiratory specimens.

Table 1: Distribution of bacterial isolates from respiratory specimens.

S. No.	Bacterial Isolates	Number of Isolates (N)	Percentage of Isolates
1	Klebsiella Pneumoniae	46	30.2
2	Escherichia coli	28	18.4
3	Pseudomonas aeruginosa	24	15.7
4	Acinetobacter species	24	15.7
5	Staphylococcus aureus	16	10.5
6	Streptococcus pneumoniae	10	6.57
7	Proteus species	04	2.63
8	Total	152	99.7

The occurrence of bacterial pathogens varies with age, in that, age group ranging from 61-70 years reported the highest number of occurrence 68 (32.3%) followed by 51-60 years 35 (16.6%). This might be probably due to the fact that, most of the people in these age groups in this community are more exposed to agents responsible for causing respiratory tract infections than the younger ones. The high prevalence of pathogens reported among patients ranging 61-70 and 51-60 years in this study, did not agree with the findings of Taura et al. 2013, in which they recorded higher occurrence of Klebsiella pneumoniae among patients ranging from 61-70 and 51-60 years. **Graph 2.**



Graph 2. Occurrence of bacterial pathogens from patients in relation to age.

Sex-related occurrence of pathogens reveals that, male subjects reported higher number of pathogens compared to their counterpart (females). This is due to more prevalent associated risk factors (e.g. smoking and chronic alcoholism) of respiratory infections in males than females. This is consistent with other studies conducted by Panda et al., 2012¹⁷ whose reported that, out of the 101 isolated organisms, 64 (63.4%) were from males while 37 (36.6%) were from females. In which in a similar study, out of 152 total isolates, 96 (63.2%) were from males while 56 (36.8%) from females. **Table:2**

Table:2 Occurrence of bacterial pathogens isolated from patients in relation to sex

Sex	No. of Patients examined	Patients with pathogens	Patients without pathogens
Male	138 (65.8%)	96 (63.2%)	42 (72.4%)
Female	72 (34.2%)	56 (36.8%)	16 (27.6%)
Total	210 (100%)	152 (100%)	58 (100%)

Table:3 Sensitivity patterns of the gram positive bacteria isolates of the respiratory tracts infections.

Bacterial Isolates	Gen tam	Cipr ofloxacin	Eryt hrom ycin	Van com ycin	Cotri moxa zole	Tetra cycli ne	Line zoli d	Ampi cillin
Streptococcus pneumoniae	++	+++	-	-	+++	-	-	-
S. aureus	++	-	-	+++	++	-	+++	-

Key:

- GEN = Gentamycin, E= Erythromycin, TE = Tetracycline, COT= Cotrimoxazole,
- CIP =Ciproflaxacin,VA= Vancomycin, LZ=Linezolid, AMP= Ampicillin
- - = resistance, ++ = Intermediate sensitive, +++ = highly sensitive

Table:4 Sensitivity patterns of gram negative bacterial isolates of the respiratory tracts infections.

Bacterial isolates	Ciproflo xacin	Cefotaxi me	Ampicillin Sulbactam	Piperacillin Tazobactam	Amikacin	Gentamy cin	Meropen em	Imipen em	Ceftazi dime	Cotrimo xazole	Aztreon am
Klebsiella pneumoniae	-	-	++	+++	++	++	++	+++	-	++	-
Escherichia coli	-	-	++	++	++	++	++	+++	-	-	-
Pseudomonas aeruginosa	++	-	-	+++	++	++	++	+++	++	-	++
Acinetobacter species	-	++	++	++	++	++	++	++	-	-	-
Proteus species	-	++	++	+++	+++	++	++	+++	-	++	-

Key:

- CIP=Ciprofloxacin, CTX= Cefotaxime, AS= Ampicillin Sulbactam, PIT= Piperacillin
- Tazobactam, AK= Amikacin, GEN= Gentamycin, MRP= Meropenem, IPM= Imipenem, CAZ= Ceftazidime, COT= Cotrimoxazole, AT= Aztreonam
- - = resistance, ++ = Intermediate sensitive/effective, +++ = highly sensitive/effective

The results from table 3 show that, *S. pneumoniae* was highly sensitive to ciproflaxacin, Cotrimoxazole, but moderately sensitive to Gentamycin. On the other hand, *S. aureus* was found to highly sensitive Vancomycin, Linezolid moderately sensitive to Gentamycin, Cotrimoxazole, and at the same time shows resistance to the remaining four antibiotics (Taura et al.2013).

The clinical relevance of these enzymes is due to their ability of causing therapeutic failures. In the present study, most of the isolates in table 4 were sensitive to gentamycin, Imipenem, Piperacillin Tazobactam, Imipenem, Ceftazidime but resistant to Cefotaxime ,cotrimoxazole, and Aztreonam. These are supported by the findings of (El- Mahmood *et al.*, 2010), whose also reported in a similar study that, the isolates were sensitive to Imipenem, Piperacillin Tazobactam, and also most were not or poorly sensitive to cotrimoxazole, Gentamycin.

CONCLUSION:

The study revealed Gram Negative Bacilli (GNB) as major pathogens causing respiratory tract infections (RTIs). *Klebsiella pneumoniae* was the predominant respiratory pathogen followed by *Escherichia coli*. Imipenem, was the most sensitive drug, next is Piperacillin- tazobactam and Amikacin and should be used for empirical therapy for RTI. The treatment should be modified as per the culture and sensitivity report from the microbiology lab.

Antibiotic resistance among respiratory bacterial pathogens is alarming. Strict implementation of the concept of 'antibiotic stewardship' has become necessary to conserve the already available antibiotics. Hospitals should have an 'antibiotic policy' and facilities for proper monitoring of antibiotic usage along with effective infection control practices to check the issue of antibiotic resistance. Weekly analysis types of respiratory pathogens and regular updation of their antibiogram should be done in every institution, so that changing trends can be identified and therapy adjusted accordingly.

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Conflict Of Interest: None declared.

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