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Original Research Article

Bacteriological profile and antibiogram of bronchoalveolar lavage fluid from patients with respiratory tract infections at a tertiary care Hospital

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ABSTRACT

Background: Chronic respiratory diseases account for 4 million deaths annually, and it is one of the most common health issues globally. The present study aims to determine bacterial isolates and their sensitivity pattern as the emergence of antibiotic resistance in the frequently isolated pathogens has complicated the use of empiric therapy with traditional agents.

Materials and Methods: BAL fluid samples of patients with respiratory infections undergoing bronchoscopy in KIMS Hospital, Amalapuram, were collected under aseptic conditions and processed according to standard protocol over 15 months, from July 2018 to October 2019. The antimicrobial susceptibility was tested by the Kirby-Bauer disc diffusion method as per CLSI guidelines.

Results: Out of 135 samples, 52(58%) were positive for growth. Of these, 49 were bacterial isolates, and 3 were fungal isolates. The highest isolation rate was observed in the 51-60 yrs age group followed by 61-70 yrs. The common organisms isolated were GNB, predominant being *Klebsiella pneumoniae* 30(61%), followed by *Pseudomonas* 15(30%) and *E.coli* 4(8%). Resistance to all cephalosporins and sensitivity to amikacin followed by piperacillin-tazobactam and other aminoglycosides were noted.

Conclusion: BAL has improved sensitivity and specificity in the diagnosis of respiratory infections. An updated antibiogram and monitoring of bacterial isolates and other susceptibility is attributed to antibiotic resistance and the changing pattern of bacterial pathogens.

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1. Introduction

Pulmonary infections are the most common Infectious disease of humans worldwide.¹ Pulmonary infections present with cough with expectoration, dyspnoea, wheeze, chest pain/ discomfort, to life-threatening infections for 1-3 weeks.² These infections pose a permanent & pervasive health problem that compose an enormous burden on society and are with reasons for consultation & hospitalization. These infections range from self-limiting illnesses to potentially life-threatening infections.³

Respiratory tract infections (RTI) are among the leading causes of morbidity & mortality in the world. RTI is a term assigned not to a single disease but a spectrum

of infections, each with different epidemiology, clinical presentation, pathology & prognosis. The etiology, signs & symptoms of respiratory diseases vary with age, sex, seasons, type of population at risk, and various other factors. The lower respiratory tract infections are responsible for 6% among all patients with infectious diseases attending out-patient departments in tertiary care hospitals and 4.4% hospital admissions.⁴

Of these, chronic respiratory diseases account for 4 million deaths annually, contributing to 5% of global deaths.¹ Exacerbations punctuate the course of the disease. Respiratory tract infections are the most frequent causes of exacerbations associated with greater & irreversible decline in lung function, significant mortality & morbidity.⁴ Early diagnosis and proper choice of antimicrobials are crucial for the management of these patients.⁵ The bacteriological

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profile of respiratory tract infections varies within the same country due to differences in the frequency of antibiotics, environmental factors, and ventilation in critically ill patients.

Since the etiological profile of respiratory infections cannot be determined clinically, the microbial investigation is required for both treatment and management of these cases.

Bronchoalveolar lavage [BAL] fluid is an ideal sample used for the recovery of these pathogens. BAL is increasingly utilized as a diagnostic tool, though it remained an investigative and research tool in the past. The advent of bronchoscopy and quantitative analysis of BAL have improved sensitivity and specificity in diagnosing pulmonary infections.

Hence, this study was performed to detect pathogenic organisms by microscopy of BAL fluid and isolate and identify various bacteria and fungi from BAL fluid in culture and analyze their antibiogram.

2. Materials and Methods

This retrospective study was conducted in the Department of Microbiology, KIMS&RF, Amalapuram, from July 2018 to October 2019. BAL fluid specimens were collected under aseptic precautions and transported to the laboratory immediately for processing. A total of 135 BAL fluid samples were included in the study. These samples were carefully observed for consistency, colour, and odour. The initial microscopic examination consisted of Gram stain and wet mount preparation, which were observed for pus cells, epithelial cells, bacteria, and yeast cells.

Later, they were inoculated on Mac Conkey Agar, Blood agar, and Chocolate agar using a sterile 4mm nichrome loop and incubated at 37° c for 24-48 hours using standard laboratory techniques. The Petri dishes were observed the next day for any bacterial growth and were identified by their morphological, cultural, and biochemical characteristics. For fungal growth, the sample was inoculated on Saborauds dextrose agar [SDA] and incubated at 37° c for seven days, and observed for growth.

The antibiotic susceptibility testing was performed by Kirby-Bauer disc diffusion method for bacterial isolates as per Clinical and Laboratory Standards Institute guideline [CLSI]-2014. Zone diameter was measured in millimetres and interpreted as per CLSI guidelines.⁵

2.1. Inclusion criteria

Patients between 20-70 years presenting with symptoms like fever with purulent sputum, breathing difficulty with physical findings suggestive of consolidation.

2.2. Exclusion criteria

Patients with cardiac diseases, and pregnant women.

3. Results

Out of the total 135 samples, 78 (58%) were from males, and 57 (42%) were female patients. The majority of cases were in the age group 51-60years, followed by the age group 61-70 yrs. The least number of patients belonged to the age group between 18-30yrs.

Out of 135 samples processed, 52 (58%) were positive for growth on culture. Most of the isolates were Gram-negative bacteria(GNB). Among 52 isolates, 49 were GNB, and 3 were fungal isolates. The predominant GNB was *Klebsiella pneumoniae* 30(61%), followed by *Pseudomonas aeruginosa* 15(30%), *Esch. coli* 4(8%) and the fungal isolate was *Aspergillus niger* 3(1%).

The highest numbers of isolates were in the age group of 51-60yrs followed by 61-70yrs, and the least number of isolates was in the age group of 18-30yrs.

3.1. Antibiotic sensitivity patterns observed

Klebsiella & *Pseudomonas* were highly sensitive to amikacin, piperacillin-tazobactam, imipenem, gentamycin, followed by tobramycin. They showed resistance to cephalosporins and slightly resistant to ciprofloxacin also.

4. Discussion

Chronic respiratory diseases represent a public health challenge in industrialized and developing countries because of their frequency and socioeconomic impact. They are the second most common cause of hospital-acquired infections with increased mortality and morbidity across the globe. Pneumonia is a frequent complication in patients with severe respiratory infections.

The present study was conducted to determine the bacterial etiology in patients with respiratory infections with the perspective of evaluating BAL fluid, which provides a handy diagnostic tool. The quality of BAL fluid was assessed based on gram stain findings followed by culture and sensitivity testing. The present study showed positive BAL cultures in 52 samples out of 135 samples collected. Of these, GNB were the predominant organisms, being 49 and 3 were fungal growths. These results are similar to the studies done by Goel et al.⁶ Barsanth et al.⁷ The maximum number of positive cases were between the age group 51-60yrs followed by 61-70yrs. This may be due to more respiratory cases observed with increasing age which lowers host defense.⁸ The rate of isolation was higher in males(58%) than in females(42%) shown in Table 1, which was on par with the findings of Shah et al.,⁹ Birasen Behera et al.¹⁰

In this study, among culture-positive GNB cases, *Klebsiella pneumoniae* was the commonest bacterial isolate followed by *Pseudomonas* and *E. coli* which is correlating with studies of Madhavi et al.,¹¹ Ratna S et al.,¹² Veena et al. and Viswanath et al.¹³

Table 1: Age and sex distribution of subjects

S.No	Age group	Male	Female	Total
1	18-30	07	05	12
2	31-40	01	16	17
3	41-50	15	06	21
4	51-60	37	14	51
5	61-70	18	16	34
Total		78	57	135

Table 2: Spectrum of bacterial isolates from BAL fluid

S.No	Isolates	No. of isolates	Percentage %
1	Klebsiella	30	61
2	Pseudomonas	15	30
3	Esch.coli	04	08
4	Aspergillus niger	03	01
Total		52	

Table 3: Occurrence of isolates in different age groups

S.No	Isolate	18-30 n =12	31-40 n =17	41-50 n =21	51-60 n =51	61-70 n =34	Total
1	Klebsiella	3(25%)	3(17.4%)	6(12%)	11(21%)	7(11%)	30
2	Pseudomonas	2(16.66%)	2(11.74%)	2(9.52%)	6(11.76%)	3(8.82%)	15
3	Esch.coli	1(6.33)	0	0	2(3.92%)	1(2.94%)	04
4	Aspergillus niger	0	0	0	2(3.92%)	1(2.94%)	03

Table 4: Resistant pattern of gram-negative isolates

Antibiotic	Klebsiella (30)	Pseudomonas (15)	E.coli (4)
Amikacin	4(13%)	3(20%)	1(25%)
Piperacillin-tazobactam	6(20%)	4(27%)	-
Gentamycin	6(20%)	4(27%)	-
Tobramycin	6(20%)	4(27%)	-
Cephalexin	25(83%)	10(67%)	3(75%)
Cefuroxime	24(80%)	12(80%)	2(50%)
Ceftriaxone	24(80%)	12(80%)	2(50%)
Cefepime	26(87%)	12(80%)	1(25%)
Ciprofloxacin	20(67%)	9(60%)	1(25%)
Tetracycline	16(53%)	10(67%)	2(50%)
Imipenem	6(20%)	3(20%)	-

In our study, all GNB isolates were susceptible to amikacin followed by piperacillin-tazobactam, gentamycin, tobramycin, and imipenem while resistant to cephalosporins. Similar findings were observed by Regha IR et al.¹⁴ Goel et al.,⁶ Barsanti et al.⁷ Olugbue V. Onouha et al.¹⁵ This resistance to cephalosporins may be due to incomplete treatment. Also, in our study Klebsiella and Pseudomonas were resistant to ciprofloxacin which is in concordance with Regha IR et al.¹⁴

In our study, most gram-negative isolates were sensitive to amikacin, piperacillin, tazobactam, followed by imipenem. Therefore it can be one of the best combinations for treating infections induced by gram-negative bacilli, which are similar to the study reported by Olugbue V. Onouha Set al.¹⁵

5. Conclusion

The study's main objective was to identify the microbiological profile of BAL fluid isolated from pulmonary infections. Results of the present study demonstrate the high incidence of gram-negative isolates. The study also suggests that regular antimicrobial sensitivity monitoring should be done as most isolates are highly resistant to cephalosporin and other commonly used antimicrobials.

6. Source of Funding

None.

7. Conflict of Interest

The authors declare that there is no conflict of interest.

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