Research Article

Bacteriological Analysis of Broncho Alveolar Wash of Patients with Suspected Pneumonia Cases

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Abstract:
Background: Respiratory tract infections are one of the commonest health issues globally. Pneumonia accounts nearly half of all deaths resulting from infectious diseases. The emergence of antibiotic resistance in the frequently isolated pathogens has complicated the use of the empiric therapy with traditional agents. The present study aims to determine the current gram negative bacterial isolates and their sensitivity pattern obtained from the bronchoalveolar lavage (BAL) fluid of patients with suspected Pneumonia cases.
Methods: Samples received from the patients attending Alluri Sita Ramaraju Academy of Medical Sciences, Eluru, were processed according to the standard protocol over a period of one year, from August 2017 to July 2018. The antimicrobial susceptibility was tested by the Kirby-Bauer disc diffusion method as per the CLSI guidelines
Results: Out of 754 samples, 121(16.04%) were culture positive for gram negative bacilli. The highest isolation rate was observed in the 41-60 years age group with a male preponderance (76.85%). The most prevalent bacterial isolates included of the present study from BAL fluid are *Klebsiella pneumoniae* (57.02 %%), *Pseudomonas aeruginosa* (29.75%). Resistance to cephalosporin's was noted. However, majority of the isolates were sensitive to carbapenems, betalactum/betalactamase inhibitors and the aminoglycosides.
Conclusions: Regular surveillance and monitoring of bacterial isolates and their susceptibility is critical owing to antibiotic resistance and the changing patterns of the bacterial pathogens.

Keywords: Gram negative bacterial isolates, Bronchoalveolar lavage, Antimicrobial susceptibility, Pneumonia.

INTRODUCTION

RTIs are a persistent and a pervasive health problem which impose an enormous burden on the society. They are common reasons for consultation and hospitalization.[1] Infections of the lower respiratory tract are responsible for 6% among all patients with infectious diseases attending outpatient departments in tertiary care hospitals and 4.4% of hospital admissions.[2] They account for 3% - 5% deaths in adult's upto the age of 60 years.[3] RTI is a term assigned not to a single disease, but to a spectrum of infections, each with a different epidemiology, clinical presentation, pathogenesis and prognosis. The etiology, signs and symptoms of respiratory diseases vary with age, sex, season, the type of population at risk and various other factors. These are commonly the first infection to occur post birth and pneumonia is quite often the final illness to occur before death.[4] In India, acute lower respiratory tract infection (ARI) alone is responsible for one million deaths. The management of lower respiratory tract infection is a challenge in terms of rational antimicrobial use, especially with range of wide array of antimicrobial agents. Additionally, the emergence of resistance to a wide range of

antibiotics has drawn attention to a need for better diagnostic techniques and development of newer drugs to allow somewhat more specific therapy,[5] it is a global concern that calls for continuing research. A number of organisms are usually implicated in their etiologies, the commonest being Gram negative bacteria,[6] followed by Gram positive organisms. Therefore, the present study was undertaken to determine the bacteriological profile and antimicrobial susceptibility pattern of isolates.

AIMS & OBJECTIVES:

The objective of the present study was to isolate and identify the gram negative bacterial etiological agents in BRONCHOALVEOLAR LAVAGE (BAL) of clinically suspected Lower Respiratory Tract Infection (LRTI) cases attending ASRAM hospital and to determine the pattern of antibiogram of the isolates which will help in the control of the infection so as to reduce the morbidity and mortality.

METHODS

The study was conducted in the Department of Microbiology,

Alluri Sitarama Raju Academy of Medical Sciences, Eluru. A total number of 754 bronchial wash (BAL) samples were collected with aseptic precautions during the study period August 2017 – July 2018.

Inclusion criteria: Patients above 20 years of age presenting with respiratory symptoms for atleast 1 week with radiological signs of pulmonary diseases were taken into account.

Exclusion criteria: Patients younger than 20 years, patients on anti-platelet drugs and anti-vitamin K medications and contaminants were excluded from this study.

The demographic details regarding age, sex, clinical diagnosis and co morbidities and viral serology (including human immunodeficiency virus, hepatitis B and hepatitis C virus) were recorded. Chest roentgenogram reports were also noted. The bronchoscopy was performed by the physician via the transnasal route using a Pentax video bronchoscope. Sample collected was transported promptly within two hours to the microbiology lab for processing.

Samples were carefully observed for consistency, specific colour & odour. Initial microscopic examination consisted of wet mount and Gram staining to observe the presence of pus cells & epithelial cells, bacteria as well as yeast cells with or without Pseudohyphae. Bronchial secretions with less than 10^3 CFU/ml were regarded as commensals or contaminants and were excluded from the study.

Samples received were inoculated on to the Blood agar, Mac Conkey agar. Plates were incubated at 37°C overnight. The next day, the growth on the Petridishes was observed for bacterial growth. The pathogens were identified by their morphologies and their cultural and biochemical characteristics according to standard laboratory procedures[7]. The antibiotic susceptibility testing was performed by Kirby-Bauer disc diffusion method as per Clinical and Laboratory Standards Institute guideline (CLSI) -2014. Zone diameter was measured in millimeters and interpreted as per CLSI guidelines[8]. Their sensitivities to Cefotaxime, Gentamycin, Ciprofloxacin,Amikacin,Ceftazidime,Imipenem, Meropenem, Tobramycin,Carbenicillin,Cefoperazone/sulbactum and Piperacillin / tazobactum were determined.

RESULTS

Out of 754 samples, 121 (16.04%) were found to be culture positive for bacterial isolates. Of those, 93 samples (76. 85%) from among males and 28 samples (23.14%) from among females were culture positive, thus showing male predominance (Figure-1).
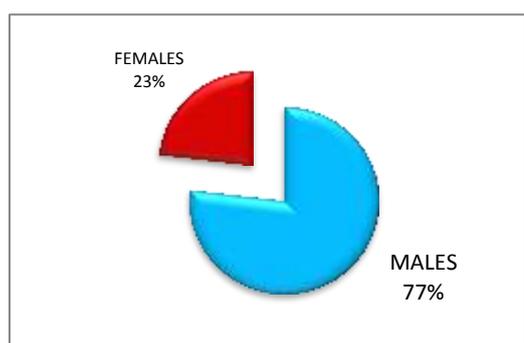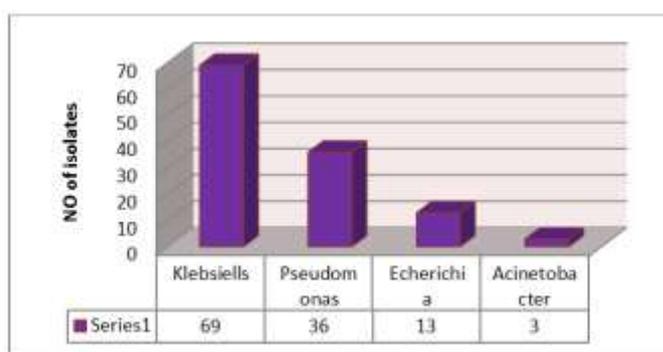

Figure 1: Sex wise distribution of gram negative isolates

All the patients above 20 years were studied. The highest isolation rate was observed in the 41-60 years age group [Table-1], followed by 61-80 age group.

Age group	Male	Female	Total
20-40	20	06	26
41-60	42	11	53
61-80	28	11	39
80 + years	03	00	03
Total	93 (76.85 %)	28 (23.14 %)	121 (100%)

Table-1: Age wise and sex wise distribution

Irrespective of the age group, among gram negative isolates *Klebsiella pneumonia* (57.02%) was found to be the predominant organism, followed by *Pseudomonas aeruginosa* (29.75%), *Escherichia coli* (10.74%), *and Acinetobacter baumannii* (2.47%) [Figure-2].

Figure-2: Distribution of Gram Negative Isolates

Klebsiella pneumonia was highly sensitive to imipenem, Meropenem, Amikacin and also to cefaperazone sulbactam. However it showed maximum resistance to ceftazidime and ciprpfloxacin. *Pseudomonas aeruginosa* showed 100% to imipenem, meropenem, followed by carbenicillin, tobramycin, Amikacin and cefaperazone sulbactam. It showed slight resistance to ceftazidime. However ceftazidime resistance was low when compared with others. *Escherichia coli* showed 100% sensitivity to carbapenems and Amikacin. However it showed high resistance to ceftazidime followed by ciprofloxacin. *Acinetobacter baumannii* showed 100% resistance to carbapenems, as well as to ciprofloxacin and ceftazidime when compared to others. However it showed resistance to Amikacin.

ANTIBIOTIC	Klebsiella pneumoniae (S%)	Pseudomonas aeruginosaa (S%)	Escherichia coli (S%)	Acinetobacter baumanii (S%)
CTX	62.31	ND	69.23	66.66
GEN	72.46	83.33	53.84	33.33
CIP	40.57	75.00	38.46	100.00
IMP	95.65	100.00	100.00	100.00
CS	84.05	86.11	76.92	66.66

CB	**ND**	97.22	**ND**	**ND**
TOB	**ND**	91.66	**ND**	**ND**
CAZ	23.18	72.22	15.38	100.00
PIT	52.17	80.55	76.92	66.66
AMI	95.65	88.88	100.00	00.00
MER	98.55	100.00	100.00	100.00

CTX = Cefotaxime   GEN = Gentamycin   CIP = Ciprofloxacin
IMP = Imipenem   CS = Cefoperazonesulbactam   CB = Carbenicillin
TOB = Tobramycin   CAZ =Ceftazidime   AMI = Amikacin
MER = Meropenem   PIT = Piperacillintazobactam   ND =Not done

Table -2: Sensitivity pattern of Gram Negative Isolates

DISCUSSION

Respiratory tract infections are the second most common cause of hospital acquired infections. An appropriate application of the clinical and laboratory findings can point towards an accurate diagnosis. Pneumonia is a frequent complication in patients with severe respiratory infection. It is frequently polymicrobial; with predominantly multi drug resistant GNB, such as *Klebsiella pneumoniae, Escherichia coli Acinetobacter baumannii, Pseudomonas aeruginosa.*[9, 10] The present study is an attempt to provide an insight on the prevalence and the antibiogram pattern of the respiratory pathogens which were isolated in a tertiary care teaching hospital of South India. Out of 754 samples, 121 (16.04%) were found to be culture positive for gram negative bacilli. Majority 53(43.80%) belong to the age group i.e., 41 to 60 years the incidence being high among male with 93 (76.85 %%) cases. *Klebsiella pneumonia* (57.02%). was the most common gram negative isolate in this study followed by *Pseudomonas aeruginosa* (29.75%)

In this study (Table -1) out of 121 culture positive cases, 53(43.80%) belong to the age group i.e., 41 to 60 years. The total incidence was found to be high among male with 93 (76.85%%) cases than in females 28 (23.14%) which was in comprehension with the findings of similar studies.[11, 12, 13] In our study, *Klebsiella pneumonia* (57.02%). was the most common bacterial isolate followed by *Pseudomonas aeruginosa* (29.75%) which was in concordance with the study.[14] *Klebsiella* showed 98% sensitive to Meropenem, followed by Amikacin (95%), Imipenem (95%), Cefaperazone Sulbactam (84%), Gentamycin (72.46%). High resistance was observed to Ceftazidime 53(76.81%), the same was reported by the study, which showed 100% resistance.[13] *Pseudomonas aeruginosa* showed 100% sensitivity to Imipenem and Meropenem followed by Carbenicillin (97%), Tobramycin (91%), Amikacin (88%), Cefoperazonesulbactum (86%). It showed slight resistance to Ciprofloxacin (27.77%) and as well as ceftazidime due to more frequent use. *Escherichia coli* showed 100% to Imipenem, Meropenem and Amikacin followed by Cefoperazone-sulbactum (76%). It showed high resistance to Ceftazidime (84.61%), Ciprofloxacin (61.53%). *Acinetobacter baumanii* showed 100% sensitivity to Imipenem, Meropenem, Ciprofloxacin and Ceftazidime. It showed 100% resistance to Amikacin. We

noticed high resistance to Cephalosporin, ceftazidime against *Escherichia coli* (84.61%), *and Klebsiella pneumoniae* (76.81%), similar observations were made by other investigators that reported 96-100% resistance.[15, 16] Fluoroquinolones are widely used in our country for different indications. Majority of the isolates were resistant to ciprofloxacin except *Acinetobacter* (0% resistant) and Pseudomonas (25% resistant). This was in correlation with the other finding.[17] In our study Meropenem sensitivity was found to be 100% for Pseudomonas, Escherichia and Acinetobacter and 98% for Klebsiella. Another study reported 100% sensitivity to meropenem against *Klebsiella* spp,[18] this finding suggests that meropenem should be used judiciously in patients to prevent any further increase in resistance. Altogether in our study majority of the isolates were sensitive to Imipenem, Meropenem and Cefoperazone-sulbactum. Majority of the isolates were sensitive to Piperacillin tazobactum, therefore it can be one of the best combination for treating infections which are induced by gram negative bacilli, as was also reported by the following study.[19] In our study Amikacin showed greater activity against majority of the isolates except *Acinetobacter baumannii* which was similar to the study made by other investogator.[20] Our study showed that majority of the isolates showed good sensitivity pattern to most of the antibiotics employed, however resistance to fluoroquinolones and 3rd generation cephalosporins was also noted .Therefore , there is a need to emphasize the prudent use of antibiotics and strictly adhere to the concept of "reserve drugs" to minimize the misuse of available antimicrobials.[21, 22]

CONCLUSION:

The present study reveals the common bacterial isolates and their sensitivity patterns. Emergence of resistant strains poses a major threat to the patients globally. Resistance to cephalosporin against predominant organisms is increasing, so need to be administered judiciously. Owing to the increased concern which surrounds antibiotic resistance and the changing patterns of bacterial pathogens, the ongoing surveillance of disease and a regular review of the management guidelines are critical. Meropenem can be effective if the isolates do not display sensitivity to other commonly used antimicrobials. The therapy should be based on the identification of at risk patients, an aggressive diagnostic work up and the broad spectrum antimicrobial treatment which is guided by microbiological support.

ethics committee

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