Original Article

Frequency of Streptococcus Pneumoniae, Haemophilus Influenzae and Moraxella Catarrhalis Causing Lower Respiratory Tract Infection in Local Population

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ABSTRACT

Objective: To see the frequency of Streptococcus pneumoniae, Haemophilus influenzae and Moraxella catarrhalis causing lower respiratory tract infection and sensitivity pattern of the isolated organisms to various antibiotics.
Study Design: Experimental Study.
Place and Duration of Study: This study was conducted at the Dept. of Microbiology Basic Medical Sciences Institute, JPMC, Karachi, from January, 2001 to September, 2001.
Materials and Methods: A total of one hundred clinically suspected cases of lower respiratory tract infections attending OPD or admitted to the wards of Jinnah Postgraduate Medical Centre (JPMC) and Civil Hospital, Karachi were included in the study.
Results: Out of 100 cases 53% cases were positive for bacterial pathogens. Of the positive cases. S. pneumonae was 35.9%, H.infuenzae 30.2% and other bacteria were 34.9%, in rest of the cases no bacterial pathogen was isolated. Age range in this study was 15-90 years and mean age was 38 years. Smokers have higher frequency i.e., 65.5% as compared to non-smokers in which 47.9% cases were positive for bacterial pathogens. Higher the number of pus cells /HPF (high power field) in sputum greater was the positivity of bacterial pathogen. Sensitivity pattern to antibiotics of different organisms was also seen in this study.
Conclusion: The goal of the study was to see the behavior of the frequent organisms on the culture and to see the antibiotic sensitivity of lower respiratory tract specimen for the treatment. It requires increased number of patients with more advanced testing system.
Key Words S.pneumonae, H.influenzae, M.cattarrhalis, lower respiratory tract infections**.**

INTRODUCTION

Respiratory tract infections (RTI) are common in our part of world due to poverty, overcrowding, poor hygienic conditions, pollution and irrational use of antibiotics. Acute RTI of viral and bacterial origin such as the common cold, pharyngitis, bronchitis, bronchiolitis, pneumonia and bronchopneumonia, pose serious problems owing to their great prevalence, associated with high mortality rates and economic costs.[1]

Common respiratory tract infections in adults in the community include acute exacerbation of chronic bronchitis (AECB), community acquired pneumonia (CAP) and acute sinusitis (AS). The most common bacterial pathogens isolated from a range of respiratory tract infections are S.pneumoniae, Haemophilus influenzae (H.influenzae) and Moraxella catarrhalis (M.catarrhalis).

Community acquired lower respiratory tract infections (LRTI) are very common and the range of causative pathogens is similar to that for community acquired pneumonia. The overall incidence is 40-50 cases per 1000 population per year.[2] Lower respiratory tract infections (LRTI) are commonly classified as either bronchitis or pneumonia, and these infections are associated with an extremely high morbidity in the community, as well as a high mortality in those patients that require hospitalization. Therefore, such infections place a huge burden, both economically and as a user of health services, on the entire health care system.[3]

The incidence of pneumonia in the developing countries is up to ten times higher than that in developed countries such as United States of America.[4] Comprehensive studies of the disease in the pre-antibiotic era showed mortality rates of about 1 per 1000 per year; over 80% of the cases were due to Streptococcus pneumoniae (S.pneumoniae).[5]

Purpose of Study: The purpose of this study was:

- To see the frequency of Streptococcus pneumoniae, Haemophilus influenzae and Moraxella catarrhalis causing lower respiratory tract infection.
- To see the sensitivity pattern of the isolated organisms to various antibiotics.
- Detection of β-lactamase production.

MATERIALS AND METHODS

This study was conducted in the Department of Microbiology, Basic Medical Sciences Institute, Jinnah

Postgraduate Medical Centre, Karachi, from January 2001 to September 2001.

Inclusion Criteria: All the patients ≥ 14 years of age suspected of lower respiratory tract infection were included in the study.

Exclusion Criteria:

- Specimen that contains only saliva.
- Patients already on antibiotic treatment.
- Patients below 14 years of age.

Early morning samples were collected in sterile containers, from the patients with clinical signs of lower respiratory tract infection. , macroscopic examination and Gram's staining and Ziehl Neelsen staining were done to see whether it was representative of lower respiratory tract infection and to exclude mycobacterium tuberculosis respectively.

The samples were inoculated onto Blood agar, Chocolate agar and on MaConkeys agar and incubated at 35° C for 24 to 48 hours suspected colonies were identified by standard methods.for the confirmation of S.pneumonia, optochin susceptibility and bile solubility and for *H.influenzae* satellitism, DNAase test and discs of X, Vand XV factor were used. β- lactamase production was tested by chromogenic cephalosporin (nitrocefin) discs.

RESULTS

A total of one hundred clinically suspected cases of lower respiratory tract infections attending OPD or admitted to the wards were included in the study.

Table No.1: Age wise Distribution of Patients with Lower Respiratory Tract Infections (n=100)

Bacterial pathogenesis	No. of cases	Percent
Positive for bacterial pathogen	53	53%
Negative for bacterial pathogen	47	47%
Age Group (Years)	Positive Bacterial Pathogen	
15 – 29 (n=53)	15	28.3
30 – 49 (n=53)	29	54.7
≥50 (n=53)	09	17.0
Age range	**15-90 years**	
Mean age	**38 years**	

Table No.2: Relationship of Smoking with Positive Culture

Smoking	No. of cases	Culture +ve		Culture –ve	
		No	%	No	%
Smoker	29	19	65.5%	10	34.5%
Non smoker	71	34	47.9%	37	52.1%

Table-1 shows the number of cases that were positive for bacterial pathogens and distribution of age, out of 100 cases 53% cases were positive, age range in this study was 15-90 years and mean age was 38 years. In 15-29 years age group, bacterial pathogens were positive in 28.3%, in age group 30-49 years bacterial pathogens were positive in 54.7%, followed by age group 50 and above in which 17% cases were positive for bacterial pathogens. Table-2 shows relationship between LRTI and smoking habits of the patients. Smokers have higher frequency i.e., 65.5% as compared to non-smokers in which 47.9% cases were positive for bacterial pathogens. Demographic data was analyzed in this table. Incidence of LRTI in different socio-economic groups was shown positive culture in lower middle class group showed a slightly higher percentage i.e., 50% as compared to other groups.

Table No.3: Demogramphic of Lower Respiratory Tract Infection in Different Social Groups

Socio-economic status	No. of cases	Culture +ve		Culture – ve		P value
		No.	%age	No.	%age	
Poor	63	33	52.3	30	47.7	0.954
Lower middle	29	16	55.2	13	44.8	
Middle	8	4	50.0	4	50.0	
Occupation						
House wife	30	13	43.3	27	56.7	0.084
Skilled worker	17	6	35.3	11	64.7	
Unskilled worker	08	04	50.0	04	50.0	
Students/ jobless/retired	13	7	53.8	6	46.2	
Patients						
Hospitalized	53	36	67.9	17	32.1	0.003
Non-hospitalized	47	17	36.2	30	63.6	

*Statistically significant

Table No.4: Frequency of Lower Respiratory Tract Infection in Normal and Compromised Respiratory Tract

Past history of respiratory illness	No. of cases	Culture +ve		Culture - ve		P value
		No	%	No	%	
Yes	17	14	82.4	03	17.6	0.017*
No	83	39	36.2	44	53.0	
Gram staining status leucocyte / HPF						
< 15	38	05	13.2	33	86.8	0.010
15 – 20	22	12	54.5	10	45.5	
> 20	40	36	90.0	04	10.0	

*Statistically significant.

Occupation had little role in causing LRTI. Incidence of bacterial pathogens in hospitalized and non-hospitalized patients was compared and was found to be more 67.9% in hospitalized as compared to 36.2% in non-hospitalized patients, (Table-3). In table-4, frequency of LRTI in cases who had no past history of respiratory disease was low i.e. 36.2% as compared to cases who had previous history of respiratory disease i.e., 84.4%. In table-4, relationship between pus cells per HPF and culture positive for bacterial pathogen was seen and it

was observed that <15 pus cells/HPF had 13.2% cases positive for bacterial pathogen. Pus cells 15-20/HPF had 54.5% cases positive for bacterial pathogen and specimens in which there were >20 pus cells/HPF, 90% cases were positive for bacterial pathogen.

Table No.5: Distribution of 53 Lower Respiratory Tract Bacterial Pathogens

Total Positive specimens	S. pneumoniae	H. influenzae	Miscellan-eous**
53	19 (35.9%)	16 (30.2%)	18 (33.9%)

****Miscellaneous LRT bacterial pathogens:**

Bacterial Isolated	No. of cases	Percent
Pseudomonas species	12	22.6%
Klebsiella pneumoniae	03	5.6%
Proteus species	01	1.9%
Staphylococcus aureus	01	1.9%
Acinetobacter	01	1.9%

Table No.6: Sensitivity Pattern of Common Lower Respiratory Tract Isolates

Antibiotics	Organism Isolated	
	Strep. Pneumoniae (n=19)	H. influenzae (n=16)
Penicillin	16 (84.2%	4 (18.7%)
Amoxycillin-clavulanic acid	19 (100%)	13 (81.2%)
Chloramphenicol	19 (100%)	14 (87.5%)
Ciprofloxacin	18 (94.7%)	15 (93.7%)
Erythromycin	10 (52.6%)	14 (87.5%)
Co-trimoxazole	3 (15.7%)	12 (75.0%)
Tetracycline	6 (31.1%)	7 (43.7%)
Ceftriaxone	19 (100%)	16 (100%)
Vancomycin	19 (100%)	16 (100%)
H. Influenzae positive	16	
β-lactanase positive	25%	

Distribution of pathogenic organisms was shown in table-5. From a total of 100 cases of LRTI, 53% were positive for pathogenic bacteria, out those 19% were due to S.pneumoniae, 16% were due to H.influenzae and 18% were due to miscellaneous bacterial pathogenic organisms. Distribution of miscellaneous bacterial pathogens was; Pseudomonas 22.6%, Klebsiella pneumoniae 5.6%, Proteus, Staph.aureus and Acinetobacter species were found to be 1% each. Sensitivity pattern of S.pneumoniae and H.influenzae was shown in Table-6. S.pneumoniae was 100% or near 100% sensitive to Ciprofloxacin, Ceftriaxone, Chloramphnicol, Vancomycin and Amoxicillin-Clavulanic acid, 84.2% sensitive to Penicillin, 31% sensitive to Tetracycline and only 15.7% sensitive to Co-trimoxazole. H.influenzae was 100% sensitive to Ceftriaxone and Vancomycin, 93.7% to Ciprofloxacin, 87.5% sensitive to Erythromycin and Chloramphenicol, 81.2% sensitive to Amoxicillin and Clavulanic acid, 75% sensitive to Co-trimoxazole, however, sensitivity to Tetracycline and Penicillin were shown to be 43.7% and 18.7% respectively. β-lactamase production in 16 cases of H.influenzae was shown and comprised of 25% cases.

DISCUSSION

Respiratory tract infections (RTI) were very common in our population; there was no current data available about the most frequent organisms which lead to wrong prescription of antibiotics, selective suppression of normal flora and emergence of resistant strains. Out of 100 clinically suspected patients with LRTI 53% had bacterial pathogens indicated that 47% of patients may not be suffering from LRTI or the etiology could be viral, fungal or of atypical bacterial origin. Several studies had shown that yield of fastidious bacteria such as S.pneumoniae and H.influenzae was zero when any specimen from respiratory tract was collected after antibiotic therapy.[6, 7] The highest incidence of LRTI was seen in the age group of 30-49 years which were the most productive years of one's life. This was in contrast to the study of Macfarlane (1993)[2] and his co-workers in which it was observed by studying 480 adult population over a period of one year; the incidence of LRTI was 2-4 times higher in people aged 60 and over. Woodhead (1995)[7] in his studies had observed that the incidence of LRTI is 15 fold higher in those over 70 years than in younger patients. This study had shown results different from international figures due to the fact that our environment is highly polluted and individuals of 30-49 years age group belonging to lower socio-economic class were exposed more to these pollutants, hence had been at greater risk to get LRTI.

Smokers were more susceptible (52.1%) to LRTI as compared to nonsmokers. These results were comparable to the study carried out by Almirall and his colleagues.[8] It was concluded from the study of 205 male and female patients that the risk of community acquired pneumonia attributable to the consumption of any type of tobacco was 32.4% of cases, in subjects without a history of COPD, the population attributable to risk of tobacco was 23%. The incidence of LRTI seen in hospitalized patients is significantly higher (67.9%) as opposed to 36.2% in non- hospitalized patients. The hospitalized patients included individuals who were clinically suspected of pneumonia or pleural effusion, chronic obstructive pulmonary disease, acute exacerbation of chronic bronchitis and asthmatic patients since they were more prone to secondary LRTI involving secondary pathogen such as Pseudomonas and Klebsiella species.[6, 9] Shaheen (1994)[10] and Sethi (2000)[11] had noted that individuals who had previous LRTI particularly in childhood have compromised respiratory tract. They could not achieve maximal lung

growth (Shaheen, 1994)[10] and were more prone to subsequent LRTI. This study also supplemented the same as 82.4% individuals with compromised respiratory tract developed infection against 36.2% of normal patients.

Gram stain can provide a basis for determining the extent to which identification and susceptibility testing of organisms recovered from specimens should be performed.[12] Sputum microscopy suggested that presence of pus cells was a good indicator for LRTI. Only 13% culture positive were seen in individuals who had < 15 pus cells/HPF as opposed to those who had > 15 pus cells/HPF 54.5%. If the pus cells are more in a sputum sample, higher is the rate of infection and subsequently LRTI, as it was reviewed by Niederman et al (1993).[13] Socio-economic status, number of siblings, maternal smoking, preterm labour, passive smoking from the atmospheric pollutants have been associated with childhood lung disease and there is failure to achieve maximal lung growth and subsequent LRTI.[14] These factors were not established in this study because the majority of the patients belonged to poor, and lower middle class.

Fang et al (1990)[15] studied 359 cases collected from multiple centers had shown that the most frequent etiologic agents were S.pneumoniae (15.3%), H.influenzae (10.9%). In 32.9% the etiology was undetermined which was comparable to this study. This study was also comparable to the study done by Macfarlane et al (1993)[2] in which out of 206 patients, 113 bacterial pathogens were isolated which included 30% S.pneumoniae, 7.7% H.influenzae, 4.3% viruses, approximately 1% cases were due to atypical pathogens. Total bacterial pathogens were accounted 41% of LRTI, whereas in this study the percentage was slightly higher. This was due to some opportunistic/secondary pathogens i.e. Pseudomonas species and Klebsiella pneumoniae. The most common pathogen in our setup was S.pneumoniae (35.9%) followed closely by H.influenzae (30%) while no M.catarrhalis was isolated which could be compared with the study of Lieberman and his co-workers (1996)[16, 17] in which etiology of community acquired pneumonia was identified in 80.6%, S.pneumoniae in 42.8%, M.pneumoniae 29.2%, C.pneumoniae 17.9%, Legionella 16.2%, Viruses 10.1%, C.burnetii 5.8% and H.influenzae 5.5% other causes were 6%.

In another study carried out by French Study Group Moine and his co-workers (1994)[18,19] found out S.pneumoniae, gram negative enterobacteriaceae and Staphylococcus aureus were commonly encountered bacterial pathogen which was in contrast to the present study, M.catarrhalis was not isolated in the present study. This might be due to shift in the pathogenicity in local set up.

It was very fortunate to know that the incidence of resistance among S. pneumoniae and H. influenzae to first line drugs such as penicillin/cephalosporin is either nil or very low. Among S.pneumoniae higher resistance is seen against Co-trimoxazole which was 84% and Tetracycline 69%. Same was true for H.influenzae as resistance to Co-trimoxazole and Tetracycline was 25% and 56% respectively .Same resistence pattern was found by Lebowitz et al in their study[20].

In the present study β-lactamase producing H.influenzae were found to be 25% which was comparable to a study carried out by Parker (1983)[10] who observed incidence of H.influenzae producing β-lactamase to be 18-22%. We must keep a constant vigilance and monitor the sensitivity pattern, so as to keep a check on resistance.

In a study carried out by Seaton and his co-workers (2000)[21] observed by studying 412 adult patients (> 15 years) in whom an episode of respiratory tract infection had been described, during which H.influenzae was isolated, were analysed. Seventy three (17.7%) isolates of H.influenzae were resistant to amoxicillin. Resistance was associated with recent hospitalization and antibiotic exposure in the community. He suggested that hospitalized patients probably received antibiotics during their admission although acquisition of the organism or the beta lactamase via plasmids from other gram negative organisms in the hospital could be a factor. An increasing number of clinical Haemophilus isolates were now resistant to penicillin.

CONCLUSION

The majority of the cases were positive for bacterial pathogens commonest was s S.pneumonae followed by H.influenza. Isolated pathogens were sensitive to Penicilins, Quinolones and erythromycin group and were resistant to Co-trimoxazole and Tetracycline. The study group was small but it suggested that the future development could be, to take a larger group with varied socio-economic status and also look for the other etiological agents(e.g. viruses), study the causes for behaviour of M.catarrhalis and also study the emergence and re-emergence of LRT pathogens.

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