

Bacterial colonization in patients with lower respiratory tract specimens: demographic profile and microbiological pattern

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Abstract

Background: *Pseudomonas aeruginosa* is an opportunistic human pathogen and is the predominant causative agent in nosocomial infections. Resistance to nearly all the available antibiotics has been shown by *P. aeruginosa*, and multidrug-resistant *P. aeruginosa* (MDRPA) is the most important source of concern in hospital-acquired infections.

Objective: To determine the prevalence of bacterial infection and the antibiotic sensitivity pattern of *P. aeruginosa* isolated from patients with lower respiratory tract infection.

Materials and Methods: This study comprised 270 patients (151 treated as inpatients and 119 as outpatients). Sputum and bronchoalveolar lavage samples were subjected to gram staining, bacterial culture, and antibiotic sensitivity for bacterial isolates as per standard techniques.

Result: The growth of pathogens was obtained from 55.9% of inpatients and 44% of outpatients. *P. aeruginosa* (25.2%) was the major organism isolated from hospitalized patients, whereas *Moraxella catarrhalis* (6.93%) was the most common pathogen isolated from outpatients. Imipenem was found to be the most effective antibiotic against *P. aeruginosa*.

Conclusion: These findings focused on careful consideration for monitoring of antimicrobial use in order to reduce the occurrence and spread of antimicrobial-resistant pathogen.

KEY WORDS: *Pseudomonas aeruginosa*, lower respiratory tract infection patients, multidrug resistant

Introduction

In general practice, the most common lower respiratory tract infections (LRTIs) were bronchitis and pneumonia, which were associated with major mortality and morbidity worldwide.^[1] On the basis of statistics, it was found that LRTIs were

found in 4.4% of hospital admissions and 6% of general practitioner consultations.^[2] The increase in the antibiotic-resistant strains of bacteria, mainly in hospitals and in the community, poses a serious issue worldwide, thereby, causing difficulty in the control measures and necessitating abundant resources and expenditure for the same.^[3]

Pseudomonas aeruginosa is a gram-negative aerobic rod, which belongs to the family *Pseudomonadaceae*. It is an opportunistic pathogen and the predominant causative agent in nosocomial infections. It is also considered as the most challenging pathogen worldwide, because of its high rate of resistance to antimicrobial agent.^[4]

P. aeruginosa is an opportunistic pathogen with inherent resistance to many antibiotics and disinfectants, including antipseudomonal penicillins, ceftazidime, carbapenems, aminoglycosides, and ciprofloxacin. Globally, the multidrug-resistant bacterial strains prevalent in hospitals and community create

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issues of due scientific concern, especially infections caused by *Pseudomonas* species and *P. aeruginosa*, in particular.

The objective of this study was to determine the prevalence of *P. aeruginosa* in LRTI patients, inpatients, and outpatients, who attended a tertiary-care hospital in Lucknow, India, and to compare their antibiotic susceptibility pattern.

Materials and Methods

This cross-sectional study was carried out in the Department of Microbiology, King George's Medical University, Lucknow, Uttar Pradesh, India, from September 2010 to August 2013. All patients admitted to the Pulmonary Medicine Ward and outpatient department (OPD) were included in this study. All the LRTI patients aged ≥ 18 years with symptoms suggestive of LRTI (i.e., two or more of the following symptoms: cough, sputum production, shortness of breath, wheeze, fever during this illness, and chest pain) who gave written consent were enrolled for the study.

The study was approved by the Institutional Ethics Committee. Sputum samples were collected from LRTI patients. In cases where patients did not produce sputum, bronchoalveolar lavage (BAL) was taken after bronchoscopy, which was done by the concerned physician.

Any patient who presented with fever, cough, expectoration, and/or dyspnea and suspected to have a LRTI as the main diagnosis, including pneumonia, exacerbation of asthma, chronic obstructive pulmonary disease (COPD), and bronchitis was included. All the samples collected (sputum and BAL) were cultured on nutrient agar, blood agar, and *Pseudomonas* isolation agar and incubated at 37°C overnight. The colonies were identified using various biochemical tests.^[5]

The antibiotic sensitivity test was performed using "Kirby–Bauer disc diffusion method." The following antibiotics were tested: gentamycin (10 µg), amikacin (30 µg), tobramycin (30 µg), ciprofloxacin (5 µg), levofloxacin (5 µg), aztreonam (30 µg), ceftazidime (30 mcg), ceftriaxone (30 µg), ampicillin (100 µg), cefepime (30 µg), piperacillin/tazobactam (100/10 µg), amoxicillin/clavulanic acid (30 µg), imipenem (10 µg), and meropenem (10 µg). Zones of inhibition were interpreted according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.^[6]

Multidrug-resistant *P. aeruginosa* (MDRPA) is defined as nonsusceptibility to at least one agent in three or more antipseudomonal categories, i.e., aminoglycosides, carbapenems, cephalosporins, fluoroquinolones, and penicillin/β-lactamase antibiotics.

Statistical Methods

The data were analyzed for mean, median, and standard deviation and *P* value (χ^2 with Yates' correction and Fisher's exact test) by using SPSS software, version 16. Statistical significance was defined when *P* < 0.05.

Result

Among the 270 respiratory samples, 202 (74.8%) of them yielded significant growth, and the rest [68 (25.1%)] of the samples were either sterile or showed contamination. A single pathogen was confirmed in 194 (72.0%) patients and eight (3.0%) revealed mixed bacterial etiology. The most common single pathogen that was isolated was *P. aeruginosa* (29.6%), followed by *Moraxella catarrhalis* (11.9%), and *Streptococcus pneumoniae* (10.3%) [Table 1].

Table 1: Distribution of bacteria pathogens among In-patients and Out-patients

Isolates	Number of cases, n (%)	Inpatients, n (%)	Outpatients, n (%)
Single pathogen			
<i>Pseudomonas aeruginosa</i>	80 (39.6)	51 (25.2)	29 (14.3)
<i>Streptococcus pneumoniae</i>	21 (10.3)	12 (5.94)	9 (4.45)
<i>Klebsiella species</i>	13 (6.43)	10 (4.95)	3 (1.48)
<i>Escherichia coli</i>	4 (1.98)	2 (0.99)	2 (0.99)
<i>Staphylococcus aureus</i>	17 (8.41)	8 (3.5)	9 (4.45)
<i>Haemophilus influenzae</i>	18 (8.91)	11 (5.44)	7 (3.46)
<i>Moraxella catarrhalis</i>	24 (11.9)	10 (4.95)	14 (6.93)
<i>Pseudomonas</i> spp	14 (6.93)	9 (4.5)	5 (2.47)
<i>Acinetobacter</i> spp	3 (1.48)	1 (0.49)	2 (0.99)
Multiple pathogens			
<i>P. aeruginosa</i> + <i>Moraxella</i>	1 (0.49)	—	1 (0.49)
<i>P. aeruginosa</i> + <i>Klebsiella</i> spp	3 (1.48)	1 (0.49)	2 (0.99)
<i>P. aeruginosa</i> + <i>Staphylococcus</i>	2 (0.99)	1 (0.49)	1 (0.49)
<i>P. aeruginosa</i> + <i>Streptococcus pneumoniae</i>	2 (0.99)	2 (0.99)	—
Total number of cases	202 (100)	118 (58.4)	84 (41.5)

Table 2: Demographic profiles for *P. aeruginosa* infection among patients attending a tertiary-care hospital

Demographic profile	No. of <i>P. aeruginosa</i> , (n = 80)	P	Total no. of patients, (n = 270)
Attended hospital as			
Inpatients	51	NS	151
Outpatients	29		119
Age (years)			
>59	48	0.002	156
<59	32		104
Gender			
Men	59	NS	215
Women	21		55
Invasive procedure			
Conducted	34	0.024	88
None	46		182
Comorbidity			
Illness	41	0.038	158
None	39		112

NS, not significant.

Table 3: Antimicrobial susceptibility pattern of *P.aeruginosa* isolates

Antibiotics	Sensitivity pattern of <i>P. aeruginosa</i> in number (%)		
	Sensitive	Resistant	Intermediate
Ampicillin	1.25	97.5	1.25
Amikacin	86.2	11.2	2.5
Amoxiclav	5	93.8	1.25
Aztreonam	47.5	50	2.5
Ciprofloxacin	57.5	42.5	—
Ceftazidime	32.5	62.5	5
Cefepime	42.5	53.8	3.8
Ceftriaxone	30	67.5	2.5
Gentamicin	2.5	95	2.5
Imipenem	90	10	—
Meropenem	72.5	23.8	3.8
Piperacillin/tazobactam	71.2	22.5	6.25
Tobramycin	70	29.6	2.5
Levofloxacin	20	78.2	2.5

Table 4: Distribution of lower respiratory tract infections by age and sex

S.No.	Age group (years)	Male subjects (N = 215)	Female subjects (N = 55)	Total, N (%)
1	18–38	39	20	59 (22.0)
2	39–58	79	26	105 (39.0)
3	>59	97	9	106 (39.2)
Total, N (%)		215 (80)	55 (20)	270 (100)

Table 5: Distribution of *P. aeruginosa* by lower respiratory tract disease

Disease of LRTI patients	N (n = 270)	Prevalence of <i>P. aeruginosa</i>	Percentage	P
COPD	139	35	25.1	0.043
Bronchitis	26	9	34.6	0.558
Pneumonia	30	9	30	0.962
Asthma	11	0	—	0.028
Lung abscess	15	6	40	0.365
Bronchitis	18	7	38.8	0.373
Carcinoma	31	14	45.1	0.044

The mean age of patients infected with *P. aeruginosa* was 51.3 or 55 years, standard deviation \pm 14.8 years (minimum 18 and maximum 85 years). The gender (male:female) ratio was 4:1, which was not significantly associated ($P = 0.093$). *P. aeruginosa* infection was significantly observed among inpatients, elderly (>59 years), associated with and had undergone any invasive procedure ($P < 0.002$) [Table 2].

The antibiotic resistance pattern is presented in Table 3. The most effective antibiotic was from carbapenem (i.e., imipenem), and its resistance rate was detected as 10%. Of the 80 samples, 52 were found to be MDRPA. There were several risk factors observed in association with the patients harboring *P. aeruginosa* including hospital stay, duration of stay, invasive device, comorbidity, and ICU stay. Significant correlation ($P = 0.031$) was observed between bacterial infections and patients with a hospital stay of more than 72 h at the time of sending the sample.

Male subjects (80%) were generally affected, and the maximum number of cases was seen among the age group 59–85 years [Table 4].

The prevalence of *P. aeruginosa* in patients with LRTI is shown in Table 5. COPD patients showed a significant association with *P. aeruginosa* ($P = 0.043$), followed by asthma ($P = 0.028$) and bronchogenic carcinoma ($P = 0.044$), which showed a significant association with *P. aeruginosa*.

Discussion

The predominance of *P. aeruginosa* resistance is considered as serious problem in many countries. It was also reported that *P. aeruginosa* is one of the most common nosocomial pathogen and a leading cause of nosocomial respiratory tract infection.^[4]

This study has demonstrated that the major single pathogens causing LRTI are *P. aeruginosa* (39.6%), *M. catarrhalis*

(11.9%), *S. pneumoniae* (10.3%), and *H. influenzae* (8.91%). These observations are different from the findings of Ozyilmaz et al.^[7] from Turkey and Liebowitz et al.^[8] from South Africa, where *H. influenzae* was the most prevalent single pathogen, followed by *S. pneumoniae* and *M. catarrhalis*. The most frequent mixed infection in this study was by *Klebsiella* species and *Pseudomonas* species (1.48%), whereas in another study by de Roux et al.,^[9] *S. pneumoniae* and *H. influenzae* constituted the most frequent cause of mixed infection. Occurrence of mixed bacterial infection in this study was 3.96%, and this is consistent with the fact that the incidence of mixed infections does not usually exceed 30%, as has been observed in other series. This study found *P. aeruginosa* as the most common gram-negative bacteria in LRTIs, as found by a study in North America,^[10] but different from another report from Japan.^[11]

In India, the prevalence rate of *P. aeruginosa* infection varies from 10.5% to 30%. It ranged from 3% to 16%, in a multicenter study conducted by Lingand Cheng.^[12]

The prevalence in our study was found to be 39.6%, which is comparable with the above-mentioned study. *P. aeruginosa* were predominantly isolated from sputum (17.4%), followed by lavage sample (12.2%). Our findings differed from those of Chaudhari et al.^[13] (35.3%) who reported that wound infection and respiratory tract infections were found to be commonly affected by *P. aeruginosa*.

Male preponderance (80%) was noted in this study. Different observations were made by Anupurba et al. (60%)^[14] and Idris et al. (57%).^[15] Outdoor activity, personal habits, nature of work, and exposure to soil, water, and other areas, which are inhabited by organism, could be the reason for male preponderance. More number of cases [106 (39.2%) cases] were seen between 59 years and 85 years, which is comparable with the studies of Anupurba et al.,^[14] in which the common age group was between 21 years and 40 years.

P. aeruginosa is inherently resistant to many antimicrobial agents, thus posing a great challenge in community-acquired and nosocomial infections. In addition, it has the ability to acquire resistance to many drug classes.

Imipenem, amikacin, and piperacillin/tazobactam were the most effective drugs observed in this study, which showed resistant rates of 10%, 11.2%, and 22.5%, respectively. Low resistance rate of *P. aeruginosa* to carbapenems and piperacillin/tazobactam may be owing to its recent introduction for use in our hospital.

Among the drugs, levofloxacin (78.2%), ceftriaxone (67.5%), and ceftazidime (62.5%) showed the highest resistance in this study. Yüce et al. (84%)^[16] and Aibinu et al. (79.4%)^[17] reported more resistance against ceftazidime in their studies. Our study is in line with the reports of Mayank et al. (63%)^[18] Ciprofloxacin showed 42.5% resistance to *P. aeruginosa* in our study. In various reports on ciprofloxacin resistance to *P. aeruginosa*, it ranged between 0% and 89%.^[19]

MDR is pervasive and growing clinical problem, which is recognized as a threat to public health in causing significance effect on morbidity and mortality and increased economic burden, which stems from the misuse of antibiotics, particularly, excessive use.

The percentage of MDR *P. aeruginosa* in India ranges from 11.36% reported by Idris et al.^[15] to 91.6% reported by Paranjothi and Dheepa.^[20] In our study, 65% *P. aeruginosa* were found to be MDR, which is comparable with above-mentioned studies.

Our study had certain limitations and strengths such as cross-sectional environment of the study with limited number of patients and period of study but we hope that it clears the way for further research. A limitation of this study could be considered that data were not available about prior antibiotic consumption of patients that might have an effect on likelihood of respiratory flora and sputum culture.

Our study has important implications. The antibiotic profile has been considered an important determinant of LRTI. This call for stringent preventive measures that includes strict infection control practices and judicious use of antibiotics implementation of antibiotic policy.

Conclusion

We can conclude that, for an effective management of LRTIs, awareness on the prevalence and antimicrobial susceptibility pattern of resistant isolates in different geographical settings would provide helpful information to the general practitioner in treatment part, which in turn has a great impact in preventing the mortality and morbidity related with *Pseudomonas* infections.

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