Original Research Article

Pattern of antibiotic sensitivity among klebsiella isolates from sputum of hospital acquired respiratory tract infection

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ABSTRACT

Klebsiella Pneumoniae is one of the most common causes of nosocomial respiratory tract infections all over the world. Unfortunately, it is developing resistance to many antibiotics. This study was carried out in a teaching hospital for determining the prevalence and pattern of antibiotic sensitivity among Klebsiella pneumoniae isolated from sputum samples of hospital acquired respiratory tract infection.

Materials and Methods: A total of 154 samples were included in the present study. Klebsiella Pneumoniae were isolated in (21.52%) samples and identified by standard microbiological techniques. Antibiotic susceptibility testing was done by the Kirby-Bauer disc diffusion method and interpreted as per CLSI guidelines.

Results: Isolated strains were sensitive to Amikacin (70%) Levofloxacin (70%) Gentamicin (65%) Chloramphenicol (65%) and resistant to Ampicillin (98.72%) Co-trimoxazole (87.5%) Ceftriaxone (85%) Tigecycline (85%) Tobramycin (85%).

Conclusion: This study may help to formulate local antibiotic policy as a part of rational antibiotic therapy for hospitals; this may reduce the development of multi-drug resistance.

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1. Introduction

Klebsiella Pneumoniae, a gram-negative opportunistic bacterium, ubiquitously present and isolated worldwide causing nosocomial infections.¹ Unfortunately it develops resistance to a various class of antibiotics including beta-lactam group. Indiscriminate prescription and overuse of beta-lactam group of antibiotics are responsible for drug resistance.² Multi-drug resistant (MDR) Klebsiella Pneumoniae isolates were defined by non-susceptibility to at least one agent in three or more antibiotic categories.³

Klebsiella Pneumoniae is a gram-negative, encapsulated, non-motile, lactose fermenting, facultative anaerobe belonging to the Enterobacteriaceae family. This was isolated by Friedlander from the lungs of patients who died of Pneumonia. The organism was named after him as Friedlander's bacillus.

Most of the Klebsiella Pneumoniae infections are associated with hospitalization mainly in immune-compromised and those with severe underlying diseases. Klebsiella Pneumoniae had been identified as a causative agent in nosocomial pneumonia (7 to 14%), septicemia (4 to 15%), wound infections (2 to 4%) and neonatal pneumonia (3 to 30%).⁴

Antibiotic resistance of Klebsiella Pneumoniae is due to spread of transmissible plasmids and acquiring resistant genes. This occurs by horizontal gene transfer that may carry virulence determinants.⁵ Capsule which is known as K antigen that protect bacteria from phagocytosis. Lipo-polysaccharide, siderophore (iron scavenging system), fimbriae (type1 & 3) are virulence factors that contribute to the pathogenicity of Klebsiella Pneumoniae. Production of ESBL by Klebsiella Pneumoniae renders resistant to many classes of antibiotics such as beta-lactam/ beta-lactamase inhibitors combinations, aminoglycosides and quinolones.⁶

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Evaluating anti-biotic susceptibility pattern of *Klebsiella Pneumoniae* which varies geographically is a must for effective control and prevention of the rapid spread of drug resistance.⁷ The prevalence of nosocomial infection due to *Klebsiella Pneumoniae* at our hospital is 21.52% and routinely beta-lactam group of antibiotics are used as a treatment. Random use of this drug may lead to MDR so to prevent and have a specific antibiotic policy of the hospital the present study of evaluating the susceptibility pattern of *K. pneumoniae* was undertaken.

2. Materials and Methods

The present study was approved by the Institutional Ethics Committee. The samples were processed in the Department of Microbiology of D.Y. Patil Medical College, Kolhapur. Sputum samples [n=158] from hospitalized patients developing respiratory tract infection were collected over a period of six months (Jan 2018 to June 2018) Written consent from volunteering participants was collected. For isolation of bacteria, the sputum samples were aseptically inoculated on blood, Mac-Conkey, and chocolate agar plates. The plates were incubated at 37 °C for 24 hours. *Klebsiella Pneumoniae* were identified by colony morphology which showed large dome-shaped colonies on blood and chocolate agar while colonies on Mac-Conkey agar were mucoid and lactose fermenting. Gram staining of sample smear revealed gram-negative, short, stout and blunt rods. Further confirmation was done by bio-chemical reactions which show indole negative, H₂S negative, V-P positive, Citrate utilization positive, Urease positive. Sugar fermentation tests exhibited production of abundant gas and acid from lactose, sucrose, glucose, mannitol and maltose.⁸

Among 158 sputum samples, 34 positive isolates were subjected to antibiotic sensitivity by Kirby Bauer disc diffusion method as per CLSI guidelines.⁹ A log-phase broth culture inoculum of isolates with turbidity equivalent to Mc-Farlands 0.5 standard (1.5 x 10⁸ CFU/ml) was prepared. It was lawn cultured on Mueller-Hinton agar and allowed to dry. The antibiotic discs were applied on the surface with sterile forceps.

Antibiotics selected were Gentamicin (10mcg), Amikacin (30mcg), Co-trimoxazole (30mcg), Tobramycin (10mcg), Aztreonam (30mcg), Ceftriaxone (30mcg), Chloramphenicol (30mcg), Nitrofurantoin (300mcg), Tetracycline (30mcg), Tigecycline (15mcg), Polymyxin B (30mcg) Levofloxacin (5mcg), Ciprofloxacin (30mcg) Piperacillin-tazobactam (100/10mcg) Amoxiclav(20/10mcg).

3. Results

Total 158 sputum samples collected during the study period were processed for culture and sensitivity. The sample population comprised of both genders. Total of 158 study

samples 34 (21.52%) have shown *Klebsiella Pneumoniae*. Gender wise 16.67% (12/72) females and 25.59% (22/86) males were positive for *Klebsiella Pneumoniae* isolation. The age group above 60 years shows the highest isolation of *Klebsiella Pneumoniae*. Among 19 females 04 (33.33%) were *Klebsiella Pneumoniae* positive & out of 25 males 09 (40.91%) were positive for *Klebsiella Pneumoniae*. Among less than 30 years of age group 8.0% (02/25) females and 13% (03/23) males were positive for *Klebsiella Pneumoniae* isolation. From Middle age group subjects 28.42% (06/28) females and 21.3% (10/38) males have shown *Klebsiella Pneumoniae* isolation.

Antibiotic susceptibility of isolates showed sensitivity to Amikacin (70%), Levofloxacin (70%), Gentamicin (65%) Chloramphenicol (65%) and were resistant to Ampicillin (98.72%), Cotrimoxazole (87.50%) Ceftriaxone (85%) Tigecycline (85%) and Tobramycin (85%).

4. Discussion

This study was carried out to find susceptibility for 16 different commonly prescribed antibiotics against *Klebsiella Pneumoniae* from Sputum samples [n=158] of hospitalized patients developing respiratory tract infections.

Present study indicates nosocomial infection due to *Klebsiella Pneumoniae* was common among male (25.59%) as compared to female (16.67%) our finding is consistent with other studies conducted by Anu Sharma¹⁰ and Gauri Kulkarni (2017)¹¹ Male preponderance may be due to the habit of smoking making them susceptible to the development of COPD.¹²

Without any gender bias individuals having more than 60 of age were found susceptible to *Klebsiella Pneumoniae* infections, this may be due to the opportunistic nature of the organism.¹³ Culture positivity of 21.52% for *Klebsiella Pneumoniae* found in our study is slightly less than Ravichitra et al⁸ and Manikandan et al¹⁴ which may be due to rational use of antibiotics in the early stage of the disease. However, our result matches with Lal et al.¹⁵

Out of 16 antibiotics tested, the present study shows a sensitivity of *Klebsiella Pneumoniae* to only a few antibiotics. Isolates were sensitive to mainly aminoglycoside and quinolones.

Due to irrational use of beta-lactam group of antibiotics for respiratory tract infections the isolates of *Klebsiella Pneumoniae* exhibited maximum resistance to them. High resistance observed for *Klebsiella Pneumoniae* to Tobramycin (85%), Piperacillin-tazobactam (71%), and Amoxiclav (70%) may be due to the production of beta-lactamase enzyme by bacteria which causes hydrolysis of beta-lactam ring, rendering them ineffective.¹⁶ Isolated *Klebsiella Pneumoniae* were sensitive to Amikacin (70%)^{15,17} Chloramphenicol (65%), Gentamicin (65%)¹⁵ these being older, less commonly prescribed antibiotics have shown good sensitivity. But routine and improper

Table 1: Frequency of age and sex wise distribution

Age in years	Females (n=72)	Culture positive in Females	Males (n=86)	Culture positive in Males
0-15	13	1(8.33%)	10	0
16-30	12	1(8.33%)	13	3 (13.64%)
31-45	11	2(16.67%)	22	7(31.82%)
46-60	17	4 (33.33%)	16	3(13.64%)
Above 60	19	4 (33.33%)	25	9(40.91%)
Total	72(45.57%)	12 (16.67%)	86(54.43%)	22(25.59%)

Table 2: Susceptibility testing for Klebsiella Pneumoniae isolates

S. No.	Antibiotic	Sensitivity (%)	Resistance (%)
1	Amikacin	70	30
2	Gentamicin	65	35
3	Aztreonam	22.5	77.5
4	Nitrofurantoin	17.5	82.5
5	Levofloxacin	70	30
6	Ciprofloxacin	20	80
7	Tobramycin	15	85
8	Polymixin B	20	80
9	Tigecycline	15	85
10	Chloramphenicol	65	35
11	Cotrimoxazole	12.5	87.5
12	Tetracycline	35	65
13	Ampicillin	1.28	98.72
14	Ceftriaxone	15	85
15	Piperacillin-tazobactam	29	71
16	Amoxiclav	30	70

exposure of isolates to newer antibiotics have shown higher resistance to them.

Isolates exhibiting a mixed reaction to Fluro-quinolones may be due to ESBL production by them which were sensitive to Levofloxacin (70%)¹⁵ while resistant to Ciprofloxacin (80%).^{10,18}

5. Conclusion

This study concludes that Amikacin, Chloramphenicol, Gentamicin and Levofloxacin antibiotics having average 65% sensitivity should be recommended under the local antibiotic policy as a part of rational antibiotic therapy for hospitals which may help to reduce the development of multi-drug resistance.

The hospital should follow uniform policy regarding the use of antibiotics which include education of patients and physicians about an appropriate drug, dose and duration of treatment. Constant surveillance of anti-microbial resistance and anti -microbial use at the hospital will help to reduce antibiotic resistance.

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None.

8. Conflict of Interest

None of the author have claimed any conflict of interest.

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