

A Study on Bacteriological Profile and Their Antibigram of Community-Acquired Pneumonia in a Tertiary Care Teaching Hospital

Koppada Rajasekhar¹

¹Associate Professor, Department of Microbiology, Guntur Medical College, Guntur, Andhra Pradesh, India.

Abstract

Background: Community-acquired pneumonia (CAP) is an acute infection of the lower respiratory tract and the most common type of lung infection that occurs outside of the hospital or within 48 hours after admission. CAP is one of the most common causes of hospitalization among infectious diseases worldwide, resulting in high fatality rates and large healthcare costs.^[1-3] **Aim:** To study the Bacteriological profile of Community-Acquired Pneumonia and their Antibigram, in patients. **Objectives:** 1. To know the most prevalent Bacterial agents associated with CAP. 2. To study the susceptibility pattern of the organisms isolated from these cases. 3. To correlate the risk factors and co-morbid conditions with the occurrence of CAP. **Subjects and Methods:** A prospective hospital-based observational study. **Study area:** Department of Microbiology, Guntur Medical College, Guntur, Andhra Pradesh. **Study Period:** 1 year. **Study population:** Suspected Community-Acquired Pneumonia patients who attended the Patient Department of General Medicine. **Sample size:** The study consisted of 500 subjects. **Sampling method:** Simple random technique. **Sample collection:** Sputum samples were collected from the patients before the initiation of antibiotics, under aseptic precautions. Early morning sputum samples were preferred whenever possible. Patients were asked to brush their teeth or rinse their mouths with water before coughing out the sputum sample. A deeply coughed-out sputum sample was collected in a sterile screw-top plastic container. A detailed history was taken, clinical parameters were noted, and Chest X-ray and routine investigations were carried out. **Results:** Streptococcus pneumoniae was 100% susceptible to Amoxicillin plus clavulanic acid and Vancomycin, followed by Cefotaxime, Azithromycin and Levofloxacin with 85.88%, 81.13% and 77.61% susceptible respectively and in ampicillin 32.96%. **Conclusion:** Pneumonia has increased in prevalence due to a lack of early detection and antibiotic resistance. The prevalence of Gram-negative bacteria as an etiological factor has also risen dramatically. According to this study, the majority of the organisms are resistant to third-generation cephalosporins. Further research should focus on how initial medication is administered in pneumonia patients.

Keywords: Community-acquired pneumonia, Pathogenic bacteria, Antibiotic susceptibility pattern.

Corresponding Author: Dr. Koppada Rajasekhar, Associate Professor, Department of Microbiology, Guntur Medical College, Guntur, Andhra Pradesh, India.

Email: rajasekharkoppada@gmail.com

Received: 26 September 2020

Revised: 16 November 2020

Accepted: 02 December 2020

Published: 30 December 2020

Introduction

Community-acquired pneumonia (CAP) is an acute infection of the lower respiratory tract and the most common type of lung infection that occurs outside of the hospital or within 48 hours after admission. CAP is one of the most common causes of hospitalization among infectious diseases worldwide, resulting in high fatality rates and large healthcare costs.^[1-3] CAP is expected to afflict 9 to 14 people per 1,000 each year, with 30 to 46% of patients requiring hospitalization.^[4-6] CAP mortality among hospitalized patients is estimated to be between 8% and 14% in the United States and 7.3% in Asian nations.^[7] Identifying and treating these people is a public health priority. Nearly 9% to 14% of all hospitalized patients required an ICU stay. According to statistics, persons requiring ICU care had a 24% mortality rate.^[8,9] The primary signs and symptoms of CAP are respiratory symptoms (cough, sputum, shortness of breath, and chest discomfort), general infection symptoms (fever, hypothermia, weakness, circulatory symptoms, and

impaired consciousness), tachypnea, tachycardia, hypotension, and focal hearing impairment. Whereas these indications are insufficiently sensitive or specific for a definitive diagnosis, confirmatory testing such as chest imaging is recommended.^[10,11]

Microorganisms have a crucial part in CAP, including the most common bacteria, Streptococcus pneumoniae, Hemophilus influenzae, Moraxella catarrhalis, and Staphylococcus aureus. These bacteria can grow in culture media and are identified using the Gram stain method. Legionella pneumophila, Mycoplasma pneumoniae, Chlamydia pneumoniae, and C. psittaci are examples of bacteria that are difficult to grow in culture conditions and cannot be stained with Gram stain.^[12,13] Despite advances in microbiological diagnostic technologies, definitive detection of CAP-causing bacteria remains a difficult task. Furthermore, collecting sufficient and uncontaminated sputum samples from the lungs is another issue that can result in false-positive results.^[5,14]

Molecular methods, such as polymerase chain reaction (PCR), have enhanced the diagnosis of many bacterial and

viral illnesses linked with CAP and have the advantage of detecting respiratory pathogens following antibiotic therapy.^[15,16] This is a prospective study to look into common causative agents of CAP, predisposing factors and sensitivity patterns of organisms that help to plan therapy.

Aim: To study the Bacteriological profile of Community-Acquired Pneumonia and their Antibiogram, in patients.

Objectives

1. To know the most prevalent Bacterial agents associated with CAP.
2. To study the susceptibility pattern of the organisms isolated from these cases.
3. To correlate the risk factors and co-morbid conditions with the occurrence of CAP

Subjects and Methods

Study Design: A prospective hospital-based observational study.

Study area: Department of Microbiology, Guntur Medical College, Guntur, Andhra Pradesh.

Study Period: 1 year.

Study population: Suspected Community-Acquired Pneumonia patients who attended the Patient Department of General Medicine.

Sample size: The study consisted of 500 subjects.

Sampling method: Simple random technique.

Inclusion criteria:

1. Age over 18 years
2. CAP was diagnosed by new or progressive pulmonary infiltrates in chest radiograph along with the presence of at least any two of the following:

- I. Fever,
- II. Cough,
- III. Purulent sputum production
- IV. Leucocytosis ($\geq 11000/\text{cumm}$)

Exclusion criteria:

Patients having tuberculosis, HIV infection, leukaemia, receiving immunosuppressive drugs, chest radiographic features suggestive of congestive cardiac failure, lung cancer, and pulmonary infarction.

Ethical consideration: Institutional Ethical committee permission was taken before the commencement of the study.

Study tools and Data collection procedure:

Sample collection: Sputum samples were collected from the patients before the initiation of antibiotics, under aseptic

precautions. Early morning sputum samples were preferred whenever possible. Patients were asked to brush their teeth or rinse their mouths with water before coughing out the sputum sample. A deeply coughed-out sputum sample was collected in a sterile screw-top plastic container. A detailed history was taken, clinical parameters were noted, and Chest X-ray and routine investigations were carried out.

Transportation: The collected sputum samples were transported to the laboratory within 2 hours of collection.

Antibiotic sensitivity testing: Antimicrobial susceptibility testing was performed by Kirby Bauer disk diffusion method on Muller Hinton agar or Mueller Hinton Agar with sheep Blood according to clinical Laboratory Standard Institute (CLSI) recommendations. Gram-negative isolates were tested for following antimicrobial antibiotic discs and the zone size interpretative chart was comparable with CLSI standards.

1. Phenotypic confirmatory disc diffusion test (PCDDT):

It is performed according to CLSI guidelines. Ceftazidime (30 µg) discs alone and in combination with clavulanic acid (ceftazidime + clavulanic acid, 30/10 µg discs) were placed on a Muller Hinton agar (MHA) plate which was inoculated with the test strain. An increase of $\geq 5\text{mm}$ in the zone of inhibition of the combination discs in comparison to the ceftazidime disc alone was considered to be an ESBL producer.

2. Double disk synergy test (disk approximation test):

MHA plate was inoculated with test strain. Augmentin (20µg amoxicillin + 10µg clavulanic acid) disc was placed in centre of the plate. Disc of cefpodoxime (30µg), ceftazidime (30µg) and cefotaxime (30µg) were placed 15-20 mm (centre to centre) around augmentin disc. Enhancement in the zone of inhibition of any cephalosporin toward augmentin indicated the presence of ESBL-producing organism production Quality control - Klebsiellapneumoniae ATCC 700603 and Escherichia coli ATCC 25922 were used as ESBL positive and negative controls, respectively.

Statistical Analysis

Data was analysed using SPSS 21.0 software. Descriptive parameters were represented as mean with SD or median. Continuous variables were compared using unpaired t-test/Mann Whitney u test. Chi-square or t-test will be used to determine significant outcome differences. Categorical data was represented as frequency with percentage. For all tests, a p-value of <0.05 was considered statistically significant.

Antibiotics	Disc content µg	Sensitivemm \geq	Intermediatemm	Resistantmm \leq
Amikacin	30	27	15-16	14
Amoxicillin/Clavulanic acid	20/10	18	14-17	13
Cefoperazone/Sulbactam	75/30	27	16-26	15
Ceftazidime	30	18	15-17	14
Ceftriaxone	30	21	14-20	13
Ciprofloxacin	5	21	16-20	15
Imipenam	10	16	14-15	13
Piperacillin/Tazobactam	100/10	21	18-20	17

Gram-positive isolates were tested for following antibiotic discs and the zone size interpretative chart was comparable with CLSI standards.

Antibiotics	Disc content µg	Sensitivemm ≥	Intermediatemm	Resistantmm ≤
Ampicillin	10	29	26-27	28
Amikacin	30	27	15-16	14
Amoxicillin/Clavulanic acid	20/10	20	-----	19
Azithromycin	15	18	14-17	13
Cefotaxime	30	23	15-22	14
Cefoxitin	30	22	-----	21
Ceftriaxone	30	21	14-20	13
Ciprofloxacin	5	21	16-20	15
Levofloxacin	5	17	14-16	13
Vancomycin	30	17	15-16	14

Klebsiella pneumonia which was resistant to generation Cephalosporins were tested for ESBL production.

Results

The study group included patients aged from 18 years to 89 years. The mean age of the patients was 51.08±15.48 (Mean ± Standard Deviation). The predominant age group was 50 – 59 years which consisted of 26.2% of cases, followed by 60 – 69 years 23%, 40-49 years 19.6%, and least 0.8 % among those below 20 years of age. More number of cases were found in patients above 50 years of age which is 58.2%

Cough and Sputum production were the most common presenting symptoms seen in 100% of cases followed by fever 89%, dyspnoea 58% and chest pain 32%. Smoking was more common at 40.6% predisposing factor among the CAP patients followed by alcoholism in 21%. In this study COPD was the commonest pre-existing condition (43%) followed by Diabetes (19%), and Cerebrovascular disorder (8%). [Table 1]

In the present study, Lobar pneumonia was the leading radiological finding in 87.2% of the CAP patients followed by Bronchopneumonia (11.8%), Interstitial pneumonia (7.4%), Pleural effusion (4.6%) and Cavitation with (1.2%). In the present study, it was observed that Leucocytosis >11,000 was found in 84.2% of cases whereas Leucopenia <4,000 was found among 6.8%. Normal levels were found in 9% of cases Raised CRP was found in 72.6% of cases. [Table 2]

Gram’s staining observation revealed the quality of sputum was suitable for diagnostic testing and bacteriological positivity was found among 44.2% of samples. Gram-positive organisms were 23% Gram negative were 21.2%. 37.2% of samples yielded bacterial growth. [Table 3]

In the distribution of the Isolates, Streptococcus

pneumoniae was the predominant pathogen at 45.69% followed by Klebsiella pneumonia at 29.03%, Pseudomonas aeruginosa at 18.27%, Staphylococcus aureus at 5.37% and Escherichia coli 1.71% of the cases. [Table 4]

Streptococcus pneumoniae was 100% susceptible to Amoxicillin plus clavulanic acid and Vancomycin, followed by Cefotaxime, Azithromycin and Levofloxacin with 85.88%, 81.13% and 77.61% susceptible respectively and in ampicillin 32.96%. [Table 6]

All the Klebsiella pneumonia and Escherichia coli isolates were 100% susceptible to Imipenem, 84.21% to Cefoperazone/Sulbactam, 78.94% to Ceftriaxone, 75.43% to Amikacin, 70.17% to Amoxicillin plus clavulanic acid, 66.66% to Ciprofloxacin, 63.16% to Cotrimoxazole and 59.64% to Ceftazidime. Among the 54 Klebsiella pneumoniae isolates, 22.22% were ESBL producers. [Table 7]

All the Pseudomonas isolates were 100 susceptible to Imipenem and Cefperazone +Sulbactum, followed by 91.17% to Piperacillin/Tazobactam, 64.7% to Ceftazidime and amikacin and 52.94% to Ciprofloxacin. [Table 8]

100 % susceptibility of Staphylococcus aureus to Imipenem, & Vancomycin, 90 % to Amoxicillin plus clavulanic acid, 80% to Ceftriaxone&Cefoxitin, 70% to Amikacin & Azithromycin, 60% to Ciprofloxacin, 30% to Ampicillin. [Table 9]

The number of patients required admission in each age group and the Mortality rates. Out of the 500 patients included in the study population, 16.2% of cases required admission and overall deaths were 5.4%. Of the number of deaths 42.8% of deaths were seen among > 80 years age and nil in < 40 years age group patients. [Table 10]

Table 1: Age and sex distribution among CAP patients

Age group in years	Total		Males		Females	
	Number	Percentage	Number	Percentage	Number	Percentage
<20	11	2.2	7	1.4	4	0.8
20-29	47	9.4	25	5	22	4.4
30-39	53	10.6	36	7.2	17	3.4
40-49	98	19.6	63	12.6	35	7
50-59	131	26.2	80	16	51	10.2
60-69	115	23	76	15.2	39	7.8
70-79	31	6.2	24	4.8	7	1.4
>80	14	2.8	8	1.6	6	1.2
	500	100	319	63.8	189	36.2

Males were 63.8% and females were 36.2% with a ratio 1: 1.69.

Table 2: Radiological findings among CAP

Radiological finding	Number of Cases	Percentage
Lobar pneumonia	436	87.2
Bronchopneumonia	59	11.8
Interstitial pneumonia	37	7.4
Pleural effusion	23	4.6
Cavitation	06	1.2

Table 3: Direct Gram staining of sputum samples

Gram staining	Number of samples	Percentage
Positive	221	44.2
Gram Positive Cocci	115	23.0
Gram Negative	106	21.2
Bacilli		
Negative	279	55.8

Table 4: Bacteriological Profile of CAP in the present study

Organism	Number isolated	Percentage
Streptococcus pneumonia	85	45.69
Klebsiella pneumonia	54	29.03
Pseudomonas aeruginosa	34	18.27
Staphylococcus aureus	10	5.37
Escherichia coli	3	1.71

Table 5: Overall Antimicrobial sensitivity

Antibiotics	Percentage
Amikacin	70.06
Ampicillin	32.96
Amoxiclav	86.72
Azithromycin	75.56
Cefotaxime	85.88
Ceftazidime	62.17
Ciprofloxacin	59.86
Ceftriaxone	79.47
Cotrimoxazole	63.16
Imipenem	100
Levofloxacin	77.61
Piperacillin+Tazobactam	91.17
Vancomycin	100

Table 6: Susceptibility pattern of Streptococcus pneumoniae (n=85)

Antibiotic	Sensitive		Resistant	
	Number	Percentage	Number	Percentage
Ampicillin	28	32.96	57	67.04
Amoxicillin/Clavulanic acid	85	100	0	0
Azithromycin	69	81.13	16	18.87
Cefotaxime	73	85.88	12	14.12
Levofloxacin	76	77.61	9	10.59
Vancomycin	85	100	0	0

Table 7: Antibiotic Susceptibility pattern of Klebsiella pneumoniae and Escherichia coli (n=57)

Antibiotic	Sensitive		Resistant	
	Number	Percentage	Number	Percentage
Amikacin	43	75.43	14	24.57
Amoxicillin/Clavulanic acid	40	70.17	17	29.83
Ceftriaxone	45	78.94	12	11.06
Ceftazidime	34	59.64	23	40.36
Cotrimoxazole	36	63.16	21	36.84
Ciprofloxacin	38	66.66	19	33.34
Cefperazone/Sulbactam	48	84.21	9	15.79
Imipenem	57	100	0	0

Table 8: Antibiotic Susceptibility pattern of Pseudomonas aeruginosa (n=34)

Antibiotic	Sensitive		Resistant	
	Number	Percentage	Number	Percentage
Amikacin	22	64.70	12	35.30
Ceftazidime	32	64.70	2	35.30

Ciprofloxacin	18	52.94	16	47.06
Cefperazone/Sulbactam	34	100	0	0
Imipenem	34	100	0	0
Piperacillin/Tazobactam	31	91.17	3	8.83

Table 9: Antibiotic Susceptibility pattern of Staphylococcus aureus (n=10)

Antibiotic	Sensitive		Resistant	
	Number	Percentage	Number	Percentage
Ampicillin	4	40	6	60
Amoxicillin/Clavulanicacid	9	90	1	10
Amikacin	7	70	3	30
Azithromycin	7	70	3	30
Cefoxitin	8	80	2	20
Ceftriaxone	8	80	2	20
Ciprofloxacin	6	60	4	40
Vancomycin	10	100	0	0
Imipenem	10	100	0	0

Table 10: Admission and mortality rates of CAP patients in the present study

Age group	Number of patients	Number of Admissions in hospital	Percentage	Deaths	Percentage
<20	11	0	0	0	0
20-29	47	1	2.12	0	0
30-39	53	3	5.66	0	0
40-49	98	10	10.20	1	1.02
50-59	131	22	16.79	5	3.81
60-69	115	25	21.73	8	6.95
70-79	31	12	38.70	7	22.5
>80	14	8	57.14	6	42.8
Total	500	81	16.2	27	5.4

Discussion

Community-acquired pneumonia, a microbial infection of the terminal airways and alveoli of the lung acquired outside of the hospital, is a devastating illness with high morbidity and death, despite the availability of powerful medications. The microbiological pattern in samples may differ, but knowledge of distribution in a specific geographical area is usually useful for initiating empirical therapy and guiding a positive clinical response.

In the present study, out of 500 cases of CAP, the microbial diagnosis was confirmed in 186 (37.2%), which was nearly correlating with Acharya et al,^[17] (39%) from Mangalore. The other studies done in India were Chintaman et al,^[18] (30.43%) from Pune, Giriraj et al,^[19] (33.8%) from Gulbarga, and Sk Jain et al,^[20] (45.5%) from Gwalior. Dheeraj Gupta et al,^[21]; pneumonia guidelines working group stated that the yield of sputum culture varies from 34% to 86%. The yield (37.2%) in the present study can be explained by the fact that the serology for both atypical and viral pathogens was not done.

The predominant age group in the present study is 50 – 59 years, where 26.2% of the patients belonged to the group. This coincides with the studies done by S K Jain et al,^[20] and Rohini et al,^[22] Bhattacharya et al,^[23] and Akter et al,^[24] reported 18 – 29 years and 21 – 30 years respectively as the predominant age groups. In the present study, 58.2% of the patients are aged >50 years which shows increasing age may be one of the risk factors for CAP.

In the present study, the Ratio of Males to females is 1.69: 1. Male preponderance is observed where males are 319

(63.8%) and females are 189(36.2%). This follows the studies of Rohini et al,^[22] Acharya et al,^[17] and Menon et al.^[25] This present study revealed 40.6% of smokers and 21% of alcoholics, nearly relating to S K Jain et al,^[20] and Rohini et al,^[22] Mucociliary clearance is defective in smokers, owing to a reduction in ciliary beat frequency and changes in volume and viscoelastic properties of respiratory secretions which leads to bacterial colonization of lower respiratory tract.

The radiological findings in the present study show a predominance of lobar pneumonia 39(78%) followed by bronchopneumonia 9 (18%), interstitial pneumonia, pleural effusion and cavitation. This study's findings relate to that of Abdullah et al,^[26] and Torres et al.^[27] Present study showed Streptococcus pneumoniae (45.7%) as the most common pathogenic organism causing CAP. It was followed by Klebsiella pneumoniae (29%) and Pseudomonas aeruginosa as the other major organisms causing CAP. Most of the studies reported Streptococcus pneumoniae as the predominant organism. Kumari et al,^[28] reported Streptococcus pneumoniae as the causative organism in 46% of CAP cases which is in concordance with the present study. Bhattacharya et al,^[23] (41.4%) are also nearly correlating with the present study. Globally, Streptococcus pneumoniae (pneumococcus) is the most common pathogen causing community-acquired pneumonia.

All the isolates in this study showed 100 % susceptibility to Carbapenems, whereas Madhulatha et al,^[29] (2013) from Bangalore reported 59.5% and Acharya et al,^[17] (2014) from Mangalore reported only 15 %, this may be due to the

rational use of Carbapenems in this region. Organisms are susceptible to Third generation Cephalosporins up to 85% in the study whereas Madhulatha et al,^[29] (2013) from Bangalore reported 73% and for 4th generation up to 83.3%, Acharya et al,^[17] (2014) from Mangalore reported up to 23% and 15 %. This study showed 100% susceptibility to Vancomycin whereas Acharya et al reported only 13 %. This study showed up to 70% susceptibility to Aminoglycosides where whereas Madhulatha et al,^[29] reported 88% and Acharya et al,^[17] up to 46%. In the present study mortality rate was 5.4% which is far below Abdulla BB et al,^[26] and Rohini et al,^[22] who reported 16, 16.5 % respectively. According to previous reports, Mortality rates range from 6-40 %. It depends upon increasing age, associated risk factors and malnutrition.

Conclusion

Pneumonia has increased in prevalence due to a lack of early detection and antibiotic resistance. The prevalence of Gram-negative bacteria as an etiological factor has also risen dramatically. According to this study, the majority of the organisms are resistant to third-generation cephalosporins. Further research should focus on how initial medication is administered in pneumonia patients.

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How to cite this article: Rajasekhar K. A study on Bacteriological profile and their Antibiogram of Community-Acquired Pneumonia in a tertiary care teaching hospital. *Asian J. Med. Res.* 2020;9(4): 11-16.

DOI: [dx.doi.org/10.47009/ajmr.2020.9.4.MB3](https://doi.org/10.47009/ajmr.2020.9.4.MB3)

Source of Support: Nil, **Conflict of Interest:** None declared.