Original Article

Clinico – Bacteriological Profile of Community-acquired Pneumonia Patients at Tertiary Care Center of North India

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Abstract

Background: Pneumonia is an acute inflammation of the pulmonary parenchyma, and its etiology can be the infective or noninfective origin. It is the sixth-leading cause of death from infectious disease in the United States, yet Indian epidemiological data were lacking on this subject. **Materials and Methods:** A hospital-based prospective observational study was done from January 2019 to June 2020. One hundred and twenty-five patients with community-acquired pneumonia (CAP) who met the inclusion and exclusion criteria during the study period were included in this study. In all patients, routine investigations and three sputum samples and two blood samples from two different sites for culture were taken on the 1st day of admission before starting the antibiotics. **Results:** Out of the 125 study participants, 80 (64%) were male, and 45 (36%) were female. The mean age of the study participants was 50.5 ± 17.2 years. Cough (99%) was the most common symptom. Chronic obstructive pulmonary disease (COPD) (25.6%) and asthma (25.6%) were the most common comorbidities. Absolute neutrophil count is the single best predictor of mortality in admitted patients of CAP (area under the curve [AUC] of 0.975) followed by total leukocyte count (AUC = 0.963) and neutrophil lymphocyte ratio (AUC = 0.925) and CRB-65 = Confusion, Respiratory rate, Blood pressure, 65 years of age and older (CRB 65 score) (AUC = 0.922) in predicting mortality in CAP. Overall bacterial growth was seen in 91 (72.8%) cases, among 74 (59.20%) Gram-negative and 17 (13.6%) were Gram-positive. **Conclusion:** *Klebsiella pneumoniae* was the most common bacteria isolated from all samples. Most of the isolates in our study were resistant to cotrimoxazole and ertapaenem, and these antibiotics should not be given as empirical antibiotics in patients of CAP.

Keywords: Absolute neutrophil count, community-acquired pneumonia, CRB-65, neutrophil-to-lymphocyte ratio ratio

Introduction

Pneumonia is an acute inflammation of the pulmonary parenchyma, and its etiology can be infective or noninfective in origin. It is the sixth-leading cause of death from infectious disease in the United States.[1] A WHO global burden of disease study revealed that lower respiratory tract illness, including community-acquired pneumonia (CAP), contributed to 429.2 million episodes of illnesses worldwide and accounted for 94.5 million disability-adjusted life years.[2] India accounts for 23% of the global burden for CAP and 36% of the regional burden for WHO.[3]

It appears that approximately 4 million cases of CAP occur annually in India, with 20% of them requiring hospitalization.[4,5] The annual incidence of CAP ranges from 5 to 11/1000 in India.[6] The mortality rate for CAP among outpatients cases is <5%, and among admitted cases, it is 10%. However, it can go beyond 30% among intensive care unit (ICU) patients.[7]

Pneumonia is increasingly common among the elderly and those with comorbidities like COPD, diabetes mellitus, renal failure, congestive heart failure, chronic lung disease, etc.[7]

Two important variables that primarily affect the spectrum of etiological agent and approach to management are the severity of initial presentation and the presence of comorbidity or advanced age.[8]

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Despite extensive diagnostic modality, the etiological agents are not detected in approximately 50% of cases of CAP.

The bacteriological profile of CAP varies geographically; therefore, it changes in different countries and within other regions of the same country. The most common etiological agent of CAP among the UK, Europe, United States, Iraq and Delhi, Shimla of India, is *Streptococcus pneumoniae*.[9-11] However, the most common etiological agent in blood culture-positive cases of CAP in Ludhiana city of India is Pseudomonas aeruginosa,[10,11,12] and in Singapore, the most common etiological agent which leads to ICU admissions is *Klebsiella pneumoniae*.[13]

Through an extensive literature review, Indian epidemiological data were lacking on this subject. This study aims to understand the clinical and bacteriological patterns of CAP at the tertiary care center of north India. This would help us better predict empirical antibiotics during exacerbations, which may differ from guidelines.

Materials and Methods

Study design

Hospital-based prospective observational study.

Study duration

January 2019 to June 2020.

Site of study

Respiratory Medicine Department.

Ethical consideration

Before initiation of the study, clearance from University Ethical Committee was taken. Before enrolment of the study, written informed consent from each patient was obtained.

Study subjects

All the patients having CAP were admitted to the Respiratory medicine department during the study duration fulfilling the inclusion and exclusion criteria.

A total of 148 patients admitted to the department of respiratory medicine were diagnosed with CAP during the study period, of which 125 patients met the inclusion and exclusion criteria.

Inclusion criteria

- Patients above the age of 18 years who have given consent for the study
- Clinically and radiologically (chest X-ray and/or CT thorax) confirmed case of community-acquired pneumonia and admitted in IPD of respiratory medicine.

Exclusion criteria

- Patients aged <18 years
- Patients who satisfy the criteria of healthcare (HOSPITAL-acquired and ventilator-associated) associated pneumonia
- Patients who are unable or unwilling to provide informed written consent for the study.

Methodology

CAP is defined as:-(a) symptoms of an acute lower respiratory tract illness (cough with or without expectoration, shortness of breath, pleuritic chest pain) for <1 week; and (b) at least one systemic feature (temperature >37.7°C, chills, and rigors, and/or severe malaise); and (c) new focal chest signs on examination (bronchial breath sounds and/or crackles), (d) Radiographic shadowing may be seen in the form of a lobar or patchy consolidation, loss of a normal diaphragmatic, cardiac or mediastinal silhouette, interstitial infiltrates, or bilateral perihilar opacities, with no other apparent cause, (e) no other explanation for the illness.[14]

Patients with the above signs and symptoms were admitted to the department of respiratory medicine, and detailed clinical history and examination of the patients were done. Then, these patients were advised for chest X-ray and/or (computed tomography [CT]) thorax (if mass or malignancy was suspected). Only clinically and radiologically confirmed patients with CAP admitted in the department of respiratory medicine were considered to study participants (after fulfilling the inclusion and exclusion criteria).

In all patients, chest radiograph, complete blood count, kidney and liver function test, serum electrolytes (Na, K, Ca), and random blood sugar along with viral markers (HIV, HBsAg, HCV) were done. In patients who cannot expectorate, sputum was induced by giving cough expectorant and nebulization with 3% normal saline and N-acetyl cysteine.

Three Sputum samples and two blood samples for culture from two different sites were taken on the 1st day of admission before initiation of the antibiotics. One sputum sample was sent to the Department of Microbiology for gram stain (GS) and culture and sensitivity (CS) along with a blood sample for culture, and a second sputum sample was sent to the DOTS (directly observed treatment short-course) center for (acid-fast bacilli [AFB]) identification. The third sputum sample was sent for sputum (cartridge-based nucleic acid amplification test [CBNAAT]) for Mycobacterium tuberculosis detection. If all the culture reports were found to be negative, video-bronchoscopy was done after taking valid written consent to take out bronchoalveolar lavage (BAL) fluid for gram staining, culture sensitivity, AFB, and CBNAAT. Trans-thoracic needle aspiration of pleural fluid was done in parapneumonic effusion and/or empyema cases. If test results of the study participants were still negative, then participants were considered as CAP without any bacterial etiology.

Special investigations

1. Chest X-ray PA view digital
2. High-Resolution CT thorax (as and when indicated)
3. Sputum for AFB, sputum samples for GS, culture, and sensitivity

4. Sputum CBNAAT for M. TB detection
5. Video-Bronchoscopy: Broncho-alveolar lavage fluid for cytology, AFB, for GS, CS and CBNAAT
6. Pleural fluid aspirate for Gram's stain and CS in case of parapneumonic effusion
7. Blood culture.

All samples were first stained by GS, then inoculated under aseptic technique on 5% blood agar and MacConkey agar plates, and were incubated aerobically at 37°C overnight. An antimicrobial susceptibility test was done by following the modified Kirby Bauer method.[15] All the collected data were entered in Microsoft Excel 2019 worksheet, SPSS software was utilized for statistical analysis. Chi-square test, Student's *t*-test, and *Z*-test were used to analyze the collected data, and $P < 0.05$ was considered significant.

Results

Out of 125 study participants, 80 (64%) were male, and 45 (36%) were female. Maximum patients (26.40%) were in the age group 60–69 years. The mean age of the study participants was 50.5 ± 17.2 years. Cough (99%) was the most common symptom. There were 42% smokers, 26.4% alcoholics, and biomass fuel smoke exposure was found in 22.4% of patients [Table 1].

COPD (25.6%) and asthma (25.6%) were the most common comorbidities among study participants. There was a history of antitubercular treatment among 28 (22.4%) patients [Table 2].

In chest X-ray unilateral (86%) and bilateral (18%), involvement was seen, and patchy consolidation (75.20%) was the most common finding. High-resolution computed tomography [HRCT] Thorax was done in 53 (42.40%) study participants. Consolidation, the most common finding on HRCT thorax, was present in 46 (86.8%) patients, followed by ground-glass opacity in 29 (54.7%) patients [Table 3].

CRB 65-scoring was used as the severity parameter of CAP among study participants. While 4 (3%) patients had zero scores, 52 (42%) patients had Score 1 on the CRB 65 scoring scale, and 54 (43%) patients scored 2. Score 3 was assigned to ten patients (8%) and Score 4–5 patients (4%). The mean CRB 65 score was 1.67 ± 0.83.

Out of 125 study participants, 94 had total leukocyte count (TLC) more than 11,000 cells/mm^3, while the remaining 31 patients had normal TLC counts. Mean TLC was 14.36 ± 5.51 cells/mm^3. When differential leukocyte count (DLC) was done, it was observed that Neutrophils were more than 70% in 121 patients (97%). Mean DLC for neutrophils was 85.46 ± 6.25%.

Neutrophil-to-lymphocyte ratio (NLR) was obtained by dividing absolute neutrophil count (ANC) with absolute lymphocyte Count. The mean value of NLR was 10.22 ± 6.9. When graph was plotted between CRB 65 score (X-axis) and NLR (Y-axis), pearson Correlation Coefficient "*r*" came out to be 0.432 while coefficient of determination

Table 1: Demographic and behavioural factors of patients

Variable name	Subgroups	*n*=125, *n* (%)
Age (years)	<30	17 (13.6)
	30-39	18 (14.4)
	40-49	18 (14.4)
	50-59	23 (18.4)
	60-69	33 (26.4)
	70-79	14 (11.2)
	>80	2 (1.6)
	Total	125
Gender	Male	80 (64)
	Female	45 (36)
Behavioural factors (risk factors)	Smokers	53 (42.4)
	Biomass fuel exposure	28 (22.4)
	Alcohol	33 (26.4)

Table 2: Clinical characteristics of community acquired pneumonia patients

Variable name	Subgroups	*n*=125, *n* (%)
Symptoms	Cough	124 (99.2)
	Sputum	112 (89.6)
	Fever	123 (98.4)
	Shortness of breath	97 (77.6)
	Chest pain	38 (30.4)
	Haemoptysis	10 (8)
	Altered sensorium	6 (4.8)
Duration of illness (weeks)	<2	49 (39.2)
	2–4	48 (38.4)
	>4	28 (22.4)
Co-morbidities	Obstructive airway disease (asthma/COPD)	32 (25.6)
	DM	11 (8.8)
	Hypertension	9 (7.2)
	AKI/CKD	6 (4.8)
	Malignancy	4 (3.2)
	CLD	3 (2.4)
	CHF/CAD	2 (1.6)
	Past history of TB	28 (22.4)

COPD: Chronic obstructive pulmonary disease, DM: Diabetes mellitus, AKI: Acute kidney injury, CKD: Chronic kidney disease, CLD: Chronic liver disease, CHF: Congestive heart failure, CAD: Coronary artery disease, TB: Tuberculosis

'r^2' was 0.187 with '$P < 0.001$, which was statistically significant [Figure 1].

The receiver operating characteristic curve (ROC) has discriminator ability of a continuous biomarker in diagnostic tests,[16] therefore sensitivity/specificity of different severity variables (CRB 65 scoring, TLC, ANC, neutrophil lymphocyte count) used for plotting the ROC Curve is the sensitivity/specificity for the prediction the mortality. Calculation of the area under the curve (AUC) was done for each variable. Moreover, it is clear from Figure 2 that Area Under ROC Curve (AUC) for ANC is highest, which means ANC is the single best predictor of mortality in CAP patients. ANC is closely followed by (TLC) with an AUC

of 0.963, making the second-best predictor of mortality in CAP and NLR (AUC = 0.925) is better than CRB 65 scoring (AUC = 0.922) in predicting mortality in patients of CAP.

Mycobacterium tuberculosis was detected in the sputum CBNAAT in 22 out of 124 sputum samples. All samples, where AFB were not seen as well as Mycobacterium tuberculosis was not detected on sputum CBNAAT ($n = 102$), were subjected to culture on Lowenstein Jensen Media where four samples showed positive growth of Mycobacterium tuberculosis. Thus, the total number of patients with pulmonary tuberculosis was 26.

In sputum, bacterial growth was seen in 84 (67.20%) and in blood, culture growth was seen in 11.20%, and in BAL, growth was seen in 06 (4.8%) in pleural fluid growth was seen in 8 (6.4%). Overall Bacterial growth was seen in 91 (72.8%) cases among 74 (59.20%) Gram-negative and 17 (13.6%) were gram-positive.

Table 3: Radiological presentation of community-acquired pneumonia patients

Variable name	Sub groups	*n*=125, *n* (%)
Chest X-ray laterality	Unilateral	107 (85.6)
	Bilateral	18 (14.4)
Chest X-ray zone involved	Right upper zone	16 (12.8)
	Right middle zone	22 (17.6)
	Right lower zone	38 (30.4)
	Left upper zone	11 (8.8)
	Left middle zone	10 (8)
	Left lower zone	40 (32)
	Whole of right lung	3 (2.4)
	Whole of left lung	1 (0.8)
Variable name	**Sub groups**	***n*=53, *n* (%)**
HRCT thorax findings	Air bronchograms	18 (34)
	Ground glass opacity	29 (54.7)
	Consolidation	46 (86.8)
	Cavity	13 (24.5)
	Parapneumonic effusion	13 (24.5)
	Tree in bud appearance	19 (35.8)
	Nodules	7 (13.2)
	Bronchiectasis	8 (15.1)
	Mass	4 (7.5)

HRCT: High-resolution computed tomography

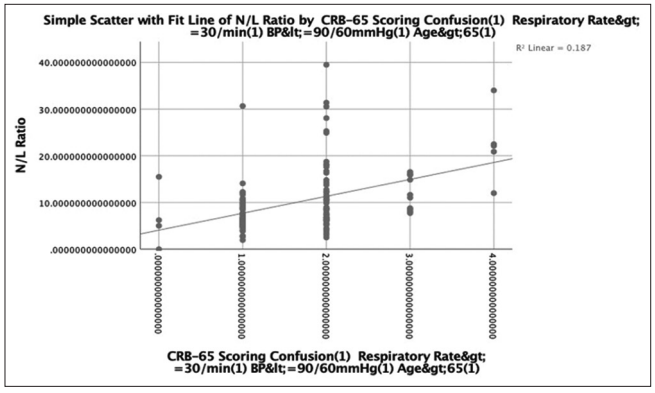

Figure 1: Graph showing a positive correlation between Neutrophil to lymphocyte and CRB-65 score

K. pneumoniae was the most common bacteria isolated from all samples, and it was 38 (35%) out of 107 isolates, followed by 16 isolates of *Escherichia coli* (15%) and 15 isolates of Pseudomonas aeruginosa (14%). K. Oxytoca grew in 2 samples (2%), while Citrobacter freundii grew in 9 samples (8%), Citrobacter koseri grew in just 1 sample (1%). Enterococcus spp and *Staphylococcus aureus* were isolated from 5 samples each (5%). *S. pneumoniae* and Acinetobacter were present in 5 samples (5%). Micrococcus and Coagulase-Negative Staphylococci were isolated from 1 sample (1%) [Table 4].

K. pneumoniae showed maximum resistance to Cotrimoxazole (72%) followed by Cefaperazone + sulbactum (48%), ceftriaxone (46%), amikacin (43%), Ertapenem (42.5%), and Amoxyclav (42%).

K. pneumoniae was most sensitive to imepenem (73.1%), Tigecycline (65%), followed by Piperacillin + Tazobactum (62.8%) levofloxacin (60%) in our study.

Pseudomonas aeruginosa showed maximum resistance against Cotrimoxazole (73%) followed by Ertapenem (67%), while it was most sensitive to Imepenem (89.4%), Meropenem (77.8%), and Levofloxacin (72.7%) [Table 5].

S. pneumoniae showed maximum resistance against cefixime (75%) while it was most sensitive to amoxycillin + clavulanic acid (100%), meropenem (100%), ertapenem (100%).

Enterococcus spp was most resistant to cotrimoxazole and ofloxacin (80% each), while it was most sensitive to amikacin (62%) and piperacillin tazatobactum (53%).

S. aureus showed maximum resistance to ciprofloxacin (80%), followed by doxycycline (71.4%), and it was most sensitive to vancomycin (100%) followed by Tigecycline (55%).

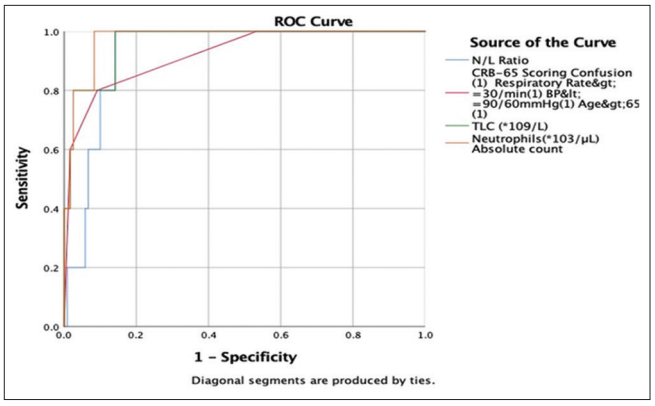

Figure 2: Plotting of receiver operating characteristic curve and measurement of respective area under curve for different scoring parameters

Coagulase-negative staphylococci were resistant to cefepime (100%), ciprofloxacin (100%) and cefoperazone + sulbactum (100%), cotrimoxazole (100%) while it was sensitive to vancomycin (100%) and amikacin (55%).

Micrococcus was resistant to cefepime (100%), cotrimoxazole (100%), cefoperazone sulbactam (100%), and gentamycin (100%) while it was sensitive to tigecycline (100%).

Discussion

In our study, the mean age of patients was 50.5 ± 17.2 years. In this study, 64% of patients were males, and 36% were females, and the most common predisposing factor among CAP patients was smoking (42%) followed by chronic Alcoholism (27%); similar observation was also reported in previous studies.[17-20]

Table 4: Distribution of bacterial growth among all samples of community-acquired pneumonia

Bacterial isolate in all samples	Count
Klebsiella pneumoniae	38
Klebsiella oxytoca	2
Escherichia coli	16
Pseudomonas aeruginosa	15
Citrobacter freundii	9
Citrobacter koseri	1
Enterococcus spp.	5
Acinetobacter spp.	4
Proteus vulgaris	1
Staphylococcus aureus	5
Streptococcus pneumoniae	4
Micrococcus	1
Nocardiaspp	1
CONS	1
Mycobacterium tuberculosis	4

CONS: Coagulase negative *staphylococcus aureus*

Neutrophil-to-lymphocyte ratio in adult community-acquired pneumonia patients

In the study done by Cataudella *et al*., they concluded that the NLR ratio was a better marker in comparison to Pneumonia Severity Index ($P < 0.05$), CURB-65, C-reactive protein, and white blood cell count ($P < 0.001$) in the prediction of 30-day mortality.[21]

Ge *et al*. found in their study that CURB-65 scores and NLR were two independent predictors correlated with unfavorable outcomes ($P < 0.05$). NLR was superior to CURB-65 in the prediction of unfavorable outcomes. They concluded that when NLR was combined with CURB-65, it had better sensitivity and specificity (89.40% vs. 91.30%).[22]

In this study, ANC was the single best predictor of mortality in admitted patients of CAP (AUC of 0.975), closely followed by TLC (AUC = 0.963). TLC was better than NLR (AUC = 0.925) and CRB 65 score (AUC = 0.922) in predicting mortality in CAP. There was a moderate positive correlation ($r = 0.432$) between CRB 65 Score and NLR with $P < 0.001$. Moreover, NLR was an independent predictor of mortality and was better than CRB 65 scoring in the prediction of mortality of CAP. So, we can conclude that ANC, TLC, and NLR are better predictors of mortality than CRB 65 score in CAP patients. They are simple, cheap, and rapidly available measurement in routine blood examination and is associated with unfavorable clinical outcomes in adult CAP patients. Thus, ANC, TLC, and NLR can be good alternatives and can be used with or without a CRB 65 score to predict mortality in CAP patients.

Table 5: Matrix for antibiotics resistance and sensitivity pattern against various bacteria

Antibiotics	*Klebsiella* (%)		*Pseudomonas* (%)		*Escherichia coli* (%)		*Citrobacter* (%)		*Acinetobacter* (%)	
	S	*R*	*S*	*R*	*S*	*R*	*S*	*R*	*S*	*R*
Ceftriaxone	54	46	66.6	33.3	-	-	-	-	50	50
Cefixime	-	-	-	-	-		50	50	-	-
Cefepime	-	-	-	-	-	-	60	40		
Ciprofloxacin	-	-	55.7	44.2	31.8	68.1	41.1	58.8	40	60
Ofloxacin	-	-	-	-	-	-	40	60	-	-
Levofloxacin	60	40	72.7	27.2	57.1	42.8	57	43	55	45
Imipenem	73.1	26.9	69	31	73.9	26.1	52.9	47.1	55	45
Meropenem	59.1	40.1.9	77.8	22.2	58.8	41.2	56.2	43.7	62.5	37.5
Ertapenem	61.5	42.5	33	67			38	62	-	-
Piperacillin + tazobactum	62.8	37.2	53	47	57.1	42.8	43	57	53	47
Cotrimoxazole	28	72	27	73	30	70	45	55	-	-
Cefoperazone + sulbactum	52	48	45	55	50	50	58.2	41.7	75	25
Amoxycillin + clavulanate	58	42	36	64	-	-	48	52	50	50
Gentamicin	-	-	-	-	-	-	-	-	-	-
Amikacin	57	43	53	47	66.6	33.3	68.4	31.6	48	52
Tigecycline	65	35	60	40	55	45	-	-	51	49
Tetracycline	-	-	40	60	43.8	55.1	-	-	55	45
Doxycycline	59.5	40.5	-	-	28.5	71.4	43	57	-	-

There is no doubt of *S. pneumoniae* being the most common cause of CAP in many studies throughout the world. However, the frequency with which it is implicated has declined and is now detected in 10%–15% of inpatient cases in the United States.[11,12,23-25]

Ibrahim *et al*. reported most frequent isolate was *K. pneumoniae* (42.8%), followed by Pseudomonas aeruginosa (30.9%), *S. aureus* (23.9%), and *E. coli* (2.4%).[26]

Chintaman *et al*. found the most common isolate *K. pneumoniae* (42.48%), followed by Pseudomonas aeruginosa (28.57%), *S. aureus* (21.4%), *S. pneumoniae* (71.4%).[25]

On the other hand, in our study on 125 patients, 73% of patients had definite bacterial etiology, out of which the most common isolate was *K. pneumoniae* present in 41.75% of patients. *E. Coli* was the second-most common bacteria isolated in 15% of samples, while Pseudomonas aeruginosa was isolated in 14% of samples was the third most common.

Mycobacterium tuberculosis was isolated on MGIT in 4 (3.7%) samples where M. TB was not detected in sputum CBNAAT. Para *et al*. showed in a study that M. tuberculosis was isolated in 4.4% of patients of CAP.[27] In previous studies, among CAP patients, M. tb was reported in approximately 5% of cases.[12,28]

Antibiotic sensitivity of bacterial isolates

Ibrahim *et al*. reported *K. pneumoniae* was resistant to third-generation cephalosporin in 16.7% of cases, and Pseudomonas aeruginosa was sensitive to Meropenem in 100%.[26]

In our study, Klebsiella pneumonia (42.8%) showed maximum resistance to Cotrimoxazole (72%) followed by Cefaperazone + sulbactam (48%), Ertapenem (42.50%), and Ceftriaxone (46%) while it was most sensitive to imipenem (73.1%), Tigecycline (65%) followed by Piperacillin Tazatobactum (62.80%).

Akter *et al*. reported that more than 80% of *S. pneumoniae* were sensitive to ampicillin, amoxycillin-clavulanate, and ceftriaxone.[29] In contrast, Gram-negative organisms were more sensitive to Meropenem, Ceftriaxone, Amoxycillin-clavulanate, and Amikacin.[29] The susceptibility to other antimicrobials ranged from 65% for azithromycin to 70% for levofloxacin.[29]

Intensive care unit admission in community-acquired pneumonia

Storms *et al*. identified 119,537 adult hospitalizations for CAP and found approximately 19% of adult pneumonia hospitalizations had an ICU admission and concluded that having a co-morbidity approximately doubled the risk of ICU admission in all age groups.[30] Our study's total number of ICU admissions was 16.8% (n = 21/125). COPD and Asthma jointly accounted for 12 out of 21 admissions (57%).

Limitations of the study

1. There are limitations of Prospective observational study as follow-up was not done, so overall mortality cannot be ascertained
2. Special test like the Urinary antigen test for Legionella spp. was not done–fastidious organisms like legionella spp. and mycoplasma spp. Do not grow on routine media, and no specific media was used to isolate them
3. Molecular tests like reverse transcription-polymerase chain reaction were not done to detect several pathogenic bacteria, including *S. pneumoniae*
4. To assess the risk factors of CAP, we need healthy individuals as control which were not present in our study as we included only hospitalized CAP patients in the study
5. Viruses are an important microbial cause of CAP, but they were not part of the study.

Conclusion

1. Gram-negative bacilli (59.20%) were found predominantly in our study, and among Gram-negative bacilli, Klebsiellapneumoniae (35%) was the most common bacterial isolate
2. ANC was the single best predictor of mortality in hospitalized CAP patients in our study, followed by TLC, NLR, and CRB-65 scoring, respectively. We recommend further study with a larger sample size to assess the importance of various markers of the predictor of mortality among CAP patients.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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