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Pneumocystis pneumonia in HIV-positive and non-HIV patients: a retrospective comparative study from a lower-middle income country

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Abstract

In this study, we compared the predisposing factors, key demographic and clinical characteristics, clinical outcomes, and factors associated with poor prognosis in pneumocystis pneumonia (PCP) infection among the human immunodeficiency virus (HIV)-positive and non-HIV patient populations. This retrospective analysis was conducted at the Aga Khan University Hospital, Karachi, via the collection and analysis of patient records with a diagnosis of "pneumocystosis" between January 2015 and October 2020. Additionally, the laboratory database was evaluated, and patients with a laboratory-confirmed diagnosis of PCP were included. During the study period, 52 laboratory-confirmed hospitalized PCP patients were identified. Of these, 23 and 29 patients were diagnosed using microscopy and polymerase chain reaction, respectively. 34.6% of our patients were HIV positive, with a median CD4 count of 20.5 cells/mm^3 (range: 10.7-50.5). Other conditions identified were corticosteroid use, autoimmune diseases, malignancy, radiation, and chemotherapy. On chest imaging, consolidation was found in 30%, ground-glass opacities in 24%, and nodular infiltrates in 20% of the cases. HIV-positive patients had a lower hemoglobin level and a higher level of β-D-glucan at the time of admission, whereas non-HIV patients were found to have more co-morbid conditions than HIV patients. We observed no difference in clinical outcomes between the two populations. Factors associated with a poor prognosis among our patients included concomitant infections at the time of diagnosis, the need for invasive mechanical ventilation, and a longer duration of stay in the hospital as well as the intensive care unit.

Key words: pneumonia, *Pneumocystis jirovecii*, HIV, acquired immunodeficiency syndrome, immunosuppression.

Introduction

Pneumocystis pneumonia (PCP), caused by *Pneumocystis jirovecii* (previously *Pneumocystis carinii)* is an opportunistic and life-threatening infection in immunocompromised individuals [1]. The affected populations include those with Human Immunodeficiency Virus (HIV), autoimmune disorders, malignancy, or patients taking immunosuppressive medications [2,3]. PCP is classified as an acquired immunodeficiency syndrome (AIDS)-defining illness for HIV-

positive individuals with a prevalence of 70-80%, and a mortality rate of 10-20% among HIV patients [3,4].

The clinical presentation of PCP differs between HIV and non-HIV-infected individuals. While HIV-positive patients tend to have a sub-acute form of illness, non-HIV patients usually have a more acute disease course, associated with a higher mortality rate [5]. Common symptoms among both sets of patients include fever, dry cough, and shortness of breath, with respiratory failure also frequently seen among non-HIV patients [5]. Diagnostic techniques for PCP include microscopy of respiratory specimens, of which bronchoalveolar lavage (BAL) and lung biopsy have higher sensitivity [6,7].

More recently, the use of polymerase chain reaction (PCR) and testing for β-D-glucan (BDG) has made diagnosis relatively easier due to its higher sensitivity and specificity [4,7]. In spite of advances in diagnostic testing, PCP continues to be a diagnostic challenge, likely owing to its non-specific signs and symptoms, use of prophylactic drugs in HIV patients, and co-infections in immunocompromised individuals [6].

Previously, low- and middle-income countries such as Pakistan were considered to be low PCP prevalent areas [8,9]. However, in recent years, there has been an increase in PCP positive patients in our hospital setting, likely due to the introduction of PCR and BDG in our laboratory. In this study, we aimed to identify the predisposing factors associated with PCP infection, compared the demographic and clinical characteristics among HIV and non-HIV patients, assessed clinical outcomes, and identified factors associated with poor prognosis in our patients.

Materials and Methods

This is a retrospective study, conducted at the Aga Khan University Hospital (AKUH), Karachi, via data collection and analysis of patient records. These records were retrieved from the Health Information Management Services at AKUH. The authors confirm that the ethical policies of the journal, as noted on the journal's author guidelines page, have been adhered to and the appropriate ethical review committee approval has been received. Approval for the study was obtained by the institute's Ethical Review Committee (ERC Number: 2019-2116-5758).

All patients having a discharge diagnosis labeled 'Pneumocystosis' between January 2015 and October 2020 were short-listed. From these, patients older than 18 years, and having a laboratory-confirmed diagnosis of PCP were included in our study. Patients who had received PCP empirical therapy but had not undergone laboratory testing were excluded. Laboratory

testing methods for PCP included direct immunofluorescence test (Bio-Rad, France) performed on sputum, BAL and lung tissue specimens until June 2018, and PCR on BAL during the latter part of the study.

A data collection sheet was prepared to collect information on demographics, pertinent past medical history (comorbid conditions, immunocompromised state, and HIV status), and details regarding hospital admission, presenting complaints, laboratory, and radiographic data, admission to the Intensive Care Unit (ICU), length of stay, treatment regimen, clinical outcome, and cause of death. Clinical charts were reviewed, and data was entered in EpiData version 6.0 (EpiData Association, Odense, Denmark).

Statistical Package for the Social Sciences (SPSS) 19 was used for data analysis. Simple frequency and descriptive analyses were run for patient demographic characteristics as well as for categorical variables, and cross-tabulation was carried out. Independent samples t-test was run to evaluate for equality of means within HIV versus non-HIV patients as well as for survivors versus non-survivors, and the p-value was calculated.

Results

We identified 52 laboratory-confirmed hospitalized PCP patients during the study period. Of these, 23 patients were diagnosed before June 2018 using direct immunofluorescence test, while the remaining 29 were diagnosed after June 2018 via PCR. The male:female ratio was 3:1. 18 (34.6%) of our patients were HIV positive, with a median CD4 count of 20.5 cells/mm 3 (range: 10.7-50.5). Among the HIV-positive patients, 6 of them had known HIV prior to their current admission for PCP while the remaining 12 patients were diagnosed with HIV at the same time as their PCP diagnosis.

Other immunosuppressive conditions identified were corticosteroid use (n=16, 30.8%), autoimmune disease (n=5, 9.6%), malignancy (n=10, 19.2%), radiation (n=3, 5.8%), and chemotherapy (n=11, 21.2%). Prior to infection, only 3 (5.8%) of our patients were receiving chemoprophylaxis for PCP. 22 (42.3%) of our patients were also found to have other concomitant infections, the majority of which were Cytomegalovirus (CMV) (n=7, 33.3%), Candidiasis (n=6, 28.6%), *Mycobacterium tuberculosis* (MTB) (n=4, 19%), and Nocardiosis (n=4, 19%).

Fever (n=26, 51%), cough (n=29, 57%), and shortness of breath (n=39, 76.5%) were found to be the most common presenting complaints. The median days for symptom duration before hospital admission was found to be 7 (range: 1-120) days. At the time of presentation, mean hemoglobin levels were 10.91±2.15 g/dL, mean platelet counts were 263±126 x 10 9 /L,

median Lactate Dehydrogenase (LDH) levels were 508 (range: 225-7958) IU/L and median BDG levels were 290 (range: 7-523) pg/mL. 42 patients also had their arterial blood gas (ABG) analysis conducted at the time of admission and a total of 18 patients (34.6%) were characterized as having acute respiratory failure (ARF) at admission based on either ABG results or clinical features. 9 of these patients had hypoxemia with PO_2<60 mm Hg, 1 patient also had concomitant hypercapnia with a PCO_2 value of 50.90 mm Hg while 2 more patients had hypercapnia with PCO_2>45 mm Hg. The remaining patients were diagnosed with ARF based on clinical features. Among patients with HIV, 6 patients (33.3%) had ARF on admission, while in those without HIV, 12 (35.3%) had ARF, thus concluding that there was no significant difference in the status of ARF among patients with and without HIV (p=0.987). Moreover, there was no significant difference between the PO_2 and PCO_2 values among patients with and without HIV (Table 1).

On chest imaging, consolidation was present in 30% (n=15), ground-glass opacities (GGOs) in 24% (n=12) and nodular infiltrates in 20% (n=10) of cases. On the other hand, reticular nodular shadowing and cavitations were each present in only 6% (n=3) of the patients. In 39 (75%) patients, the findings on chest imaging were present bilaterally. 19 (36.5%) patients were put on non-invasive ventilatory (NIV) support while 14 (27%) underwent endotracheal intubation and another 2 (3.8%) patients underwent tracheostomy. Among patients with HIV, 6 had to be put on NIV and another 6 patients were put on invasive ventilatory support. 45 (88%) patients received adjuvant steroid use. All patients were treated with a combination regimen of trimethoprim/sulfamethoxazole for 21 days. Additionally, 16 patients also received antibiotic therapy. 16 (30.8%) of our patients expired, while the rest were eventually discharged. Table 2 represents the patients' baseline and in-hospital characteristics.

We compared the characteristics between our HIV and non-HIV patients and the results are presented in Table 1. Patients with HIV were significantly younger at the time of PCP diagnosis (p<0.005). HIV-positive patients also had a lower hemoglobin level (p=0.03) and a higher level of BDG (p=0.00). It was noted that non-HIV patients with PCP were more commonly found to be diabetic (p=0.04) and hypertensive (p=0.04) as compared to the HIV group. There were four patients who had Coronavirus Disease 2019 (COVID-19) infection prior to PCP, all of whom were HIV negative. We did not observe any significant difference between the duration of symptoms before presentation between HIV and non-HIV patients (14 vs 23 days, respectively). Both groups had a similar mean length of hospital stay of 11 days, while the mean length of ICU stay was also similar (2 days in HIV vs 3 days in non-HIV) with no statistically significant difference.

Among our patients, a significant relationship was observed between the need for invasive ventilation and in-hospital mortality, with a 62.5% mortality rate among patients who underwent either endotracheal intubation, tracheostomy, or both (p=0.001). Moreover, concomitant infection at the time of diagnosis (p=0.049), bilateral findings on chest imaging (p=0.037), and the need for ICU stay (p=0.000) were also significantly associated with in-hospital mortality among patients. Increased length of ICU and hospital stay were also significantly associated with mortality (p=0.017 and p=0.022, respectively).

Discussion

In recent years, Pakistan has experienced a rise in HIV cases, likely owing to more spread among pre-existing high-risk groups along with a surge in concentrated HIV epidemics-[10,11]. Moreover, with the advent of National AIDS Control Program (NACP) in Pakistan, an increasing number of HIV cases are also now being registered. Since HIV is a strong predisposing factor for PCP infection, it may have led to an increase in PCP cases in recent years. Additionally, a change in laboratory testing methods may also play a role in the increased detection of PCP cases. The use of BAL for PCR has a higher sensitivity when diagnosing cases of PCP as compared to microscopy of sputum which has a lower yield [12,13]. In this study, we identified 52 laboratory-confirmed patients with PCP from 2015 till 2020, whereas we previously reported 37 cases from 1995 till 2015, out of which only 33% were laboratory-confirmed PCP [3]. The majority of the patients in our study were diagnosed via PCR on BAL samples.

Consistent with our observation, Bienvenu et al. reported a male predominance among their cohort of patients with PCP [14]. 34.6% of our patients were HIV positive while the remaining 65.4% had other predisposing risk factors such as autoimmune disease, corticosteroid use, or malignancy. Interestingly, four of our patients also had COVID-19 infection prior to the PCP episode. The percentage of HIV-associated PCP observed in our patients has increased compared to previous reports from Pakistan [3]. A study from Taiwan reported 23% of their patients to be HIV positive, which is less than what was observed in our findings [15]. The same study described corticosteroid use as a major risk factor among 62.9% of their subjects. In our study, 88% of the patients were taking corticosteroids. Additionally, our cohort of patients with HIV was found to be significantly younger at the time of diagnosis of PCP infection. This is similar to the findings reported by Liu et al. with a mean age of 34 years among HIV-infected patients and 60 years among non-HIV-infected patients [15]. In our study,

the mean age of HIV patients was found to be 40 years and that of non-HIV patients was 54 years.

Previous studies have reported differences between HIV and non-HIV patients in the duration of symptoms, hospital and ICU stay, and clinical outcomes [15,16]. However, in our study, there was no significant difference between these variables. One of the reasons for this could be the low sample size of our study. Diagnosis of PCP in non-HIV patients was supported by the clinical (immunocompromised state other than HIV), radiological and other ancillary data like raised LDH. Many of our non-HIV patients had a negative BDG and may have PCP colonization only versus infection. GGOs have been described as the characteristic chest CT image finding among patients with PCP [4]. However, in our cohort of patients, GGOs were found in 24% of patients whereas consolidation was the predominant finding in approximately 30% of patients. Among our HIV-positive patients, 11.1% had GGO, while consolidation and patchy shadowing were present in 33.3% and 27.8% of patients, respectively. Our findings are in contrast with the literature as GGOs are present in majority of the cases whereas consolidation is an uncommon finding [17,18]. In our non-HIV cohort, GGOs were present in 31.3% of the patients which is consistent with other studies [19].

Our patients were managed with combination therapy of trimethoprim sulfamethoxazole. Some patients also required additional treatment with antibiotics and antifungals. Approximately 88% of the patients received adjuvant corticosteroid therapy. The role of adjuvant steroid in the treatment of PCP is conflicting. Studies have suggested the role of adjuvant corticosteroid as an anti-inflammatory agent in conjunction with standard PCP therapy. Moreover, adjuvant corticosteroid use has been shown to reduce mortality [20]. However, another recent study has reported no benefit of adding early corticosteroids to PCP therapy in non-HIV patients [21].

A recent meta-analysis reported better clinical outcomes with corticosteroids only in PCP patients with respiratory failure. This study also advised against use of corticosteroids in non-HIV PCP patients without low oxygen saturation [22].

PCP is associated with a high mortality rate as a meta-analysis reports a mortality rate of 30.6% in non- HIV PCP patients [23]. Liu et al. reported higher a mortality rate of 46.4% in non-HIV patients compared to 16% HIV patients [15]. In our study, we found a higher mortality rate of 38.9% among our cohort of HIV patients, as compared to a comparatively lower mortality rate of 26.5% among the non-HIV patients. The most common cause of death in our patients was identified as ARF which is also consistent with the findings of Liu et al [20]. Although the use of NIV is shown to reduce mortality in patients with HIV who develop severe ARF due to PCP

[24], as well as among other critically ill patients with ARF [25], we observed no such association as the use of invasive and non-invasive ventilatory support was significantly associated with a higher mortality among our patients. This may be due to the fact that patients who developed severe ARF clinically deteriorated leading to eventual death. Alternatively, among our patients with ARF, not everyone received ventilatory support which may also have contributed to a higher mortality rate among those with ARF.

Our study had few limitations. This was a single-center study due to which our sample size was limited. Moreover, the retrospective nature of the study was also a limitation. Additionally, since HIV-positive patients in Pakistan receive treatment via the National Aids Control Programme (NACP), which is a Government-run initiative, there was no information available in hospital records about their HIV treatment status. Lastly, since PCP is an infrequent cause of pneumonia, especially in Pakistan, there is no standardized treatment being followed nor were all of our immunocompromised patients receiving prophylaxis for PCP infection. Because clinical suspicion needs to be high for physicians to test patients for PCP, it is possible that not every patient with the disease was tested for it, thus leading to a small sample size in our study. These factors highlights a significant gap in clinical practice which must be addressed, given the high mortality rate of PCP.

Conclusions

In recent years, an increase in the number of PCP cases has been reported, likely owing to the higher sensitivity of PCR which led to identification of more cases of PCP along with an increase in the number of HIV cases in Pakistan. PCP is associated with a high mortality rate of 30.8% in our study. Some of the factors associated with a poor prognosis seen among our patients include the need for invasive ventilation, ICU stay, and a prolonged hospital stay. There was no significant difference seen in clinical outcomes in our cohort of HIV and non-HIV patients.

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Table 1. Comparison of human immunodeficiency virus (HIV) and non-HIV patients (n=52).

	HIV (n=18)	Non-HIV (n=34)	p value
Age, years, mean±SD	39.6 ± 14.8	53.9 ± 17.6	0.005
Gender, N (%)			
Male	13 (72.2)	26 (76.5)	0.73
Female	5 (27.8)	8 (23.5)	
Laboratory parameters, mean±SD			
Hemoglobin, g/dL	10.1±1.9	11.4±2.2	0.03
BDG, pg/mL	460.1±152.6	200.1±180.2	0.00
PO_2, mm Hg	89.28±38.25	83.41±37.26	0.62
PCO_2, mm Hg	32.07±8.08	32.30±9.19	0.93
Comorbidities, N (%)			
Diabetes Mellitus	2 (11.1)	13 (38.2)	0.04
Hypertension	2 (11.1)	13 (38.2)	0.04
Concomitant infection, N (%)	10 (55.6)	12 (35.3)	0.15
Candidiasis	3	3	
Nocardiasis	0	4	
CMV	4	3	
MTB	3	1	
Aspergillus species	0	3	
Corticosteroids use, N (%)	1 (5.6)	15 (44.1)	0.00
COVID-19, N (%)	0	4 (11.8)	0.23
Chest imaging, N (%)			
Consolidation	6 (33.3)	9 (28.1)	
GGOs	2 (11.1)	10 (31.3)	
Patchy shadowing	5 (27.8)	2 (6.3)	
Pleural effusion	2 (11.1)	7 (21.9)	
Nodular infiltrates	2 (11.1)	8 (25.0)	
Outcome, N (%)			
Discharge	11 (61.1)	25 (73.5)	
Death	7 (38.9)	9 (26.5)	0.36

HIV, human immunodeficiency virus; SD, standard deviation; BDG, β-D-Glucan; PO_2, partial pressure of Oxygen; PCO_2, partial pressure of carbon dioxide; CMV, cytomegalovirus; MTB, mycobacterium tuberculosis; COVID-19, coronavirus disease 2019.

Table 2. Baseline characteristics of patients with pneumocystis pneumonia (n=52).

Variable	N (%) or mean±SD
Age, years	49±17.9
Gender	
Male	39 (75)
Female	13 (25)
Comorbidities	
Hypertension	15 (28.8)
Diabetes Mellitus	15 (28.8)
ILD*	3 (5.8)
Chronic Kidney Disease	3 (5.8)
Hypothyroidism	2 (3.8)
Ischemic Heart Disease	2 (3.8)
HIV-positive	18 (34.6)
Malignancy	10 (19.2)
Hematological	5 (9.6)
Acute Myeloid Leukemia	1 (1.9)
Diffuse large B cell Lymphoma	1 (1.9)
Hodgkin Lymphoma	1 (1.9)
Primary Central Nervous System Lymphoma	1 (1.9)
T cell Lymphoma	1 (1.9)
Solid Organ	5 (9.6)
Breast carcinoma	2 (3.8)
Neoplasia of lung	2 (3.8)
Glioblastoma Multiforme Grade 4	1 (1.9)
Autoimmune disease	5 (9.6)
Autoimmune Hemolytic Anemia	1 (1.9)
Autoimmune Hepatitis	1 (1.9)
IgA Nephropathy	1 (1.9)
Protein S deficiency	1 (1.9)
Sarcoidosis	1 (1.9)
Concomitant infections	22 (42.3)
Adjuvant steroid use	46 (88)
Complications	
ARF	18 (34.6)
Septic shock	6 (11.5)
Pneumothorax	3 (5.8)
ILD exacerbation	2 (3.8)
Outcome	
Discharge	33 (63.5)
Death	16 (30.8)
Cause of death	
ARF	8 (50.0)
Cardiopulmonary arrest	4 (25.0)
Multi-organ failure	4 (25.0)

ARF, acute respiratory failure; ILD, interstitial lung disease