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Prevalence and clinical impact of bacterial co-infection in chronic pulmonary aspergillosis

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Abstract

Background The clinical significance of co-infection with chronic pulmonary aspergillosis (CPA) and bacteria is limited and has mostly been studied in specific patient groups. This study aims to investigate the incidence and prognostic impact of bacterial co-infection in patients with CPA.

Methods A single-center, retrospective, observational study was conducted between 2019 and 2024. Patients were categorized based on the presence of bacterial co-infection, and their demographics, potential underlying factors, and prognosis were analyzed.

Results A total of 101 patients were included (mean age: 57 ± 13 years, 79 male). Bacterial co-infection was identified in 21 patients (21%). The most common bacterial pathogens at diagnosis were *Pseudomonas aeruginosa* (*n* = 6), *Klebsiella pneumoniae* (*n* = 5), *Escherichia coli* (*n* = 4), and *Serratia marcescens* (*n* = 4). Five patients had a history of prior bacterial colonization. At diagnosis, more than one bacterial species were identified in six patients. Sputum production and hypoxemic respiratory failure were more frequently observed in patients with bacterial co-infection. Systemic corticosteroid use was more common in the co-infected group. However, radiological findings and diagnostic procedures did not differ between the groups. Surgical interventions were more commonly performed in the non-co-infected group. During the follow-up, hospital admission rates, mortality, and overall survival were comparable between the two groups.

Conclusions Bacterial co-infections are probable in CPA and follow-up results of both patient Groups may not differ. Timely diagnosis and close follow-up of these patients are probable key factors in these patients.

Keywords Aspergillosis, *Klebsiella pneumoniae*, Pneumonia, *Pseudomonas aeruginosa*

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Background

In recent years, the incidence of fungal diseases has been on rise due to increased clinical awareness, advancements in imaging techniques and novel and easily-accessed diagnostic tests. The global annual incidence of chronic pulmonary aspergillosis (CPA), considering only patients with a history of tuberculosis (TB), is reported to be 1,837,272 [1]. These numbers are believed to be an underestimation, as CPA commands a wide spectrum of underlying lung diseases such as chronic obstructive pulmonary disease (COPD), sarcoidosis, and pneumothorax. Notably, approximately 2.6% of patients with progressive lung cancer and 2.5% of hospitalized COPD patients are estimated to develop invasive aspergillosis [2]. The documented mortality of chronic aspergillosis are 8% in treated patients and 20% in untreated cases [1].

Aspergillus spp. may coexist with a diverse variety of bacteria. The coexistence of *Stenotrophomonas maltophilia* and *Pseudomonas aeruginosa (P. aeruginosa)* and *Aspergillus* spp. has been widely reported [3, 4]. However, studies on such co-infections are often limited in scope, primarily consisting of case reports or research focused on specific patient groups.

The rate of bacterial and fungal co-infection has been reported as 8% in COVID-19 patients [5]. In cystic fibrosis (CF), bacterial infections are considered a risk factor for the development of fungal infections [6]. The co-colonization rate of *Aspergillus fumigatus* and *P. aeruginosa* in CF patients has been reported as 3.1% [7], with co-infected patients demonstrating an increased risk of exacerbations and hospitalizations [3, 7]. Additionally, a recent report indicated co-infection involving *Klebsiella pneumoniae (K. pneumoniae)* and *Aspergillus fumigatus* [8].

Bronchiectasis may accompany chronic bacterial infections, often presenting with recurrent exacerbations [9]. The most commonly isolated bacteria in these patients include *Haemophilus influenzae*, *P. aeruginosa*, *Streptococcus pneumoniae*, *Staphylococcus aureus*, and *Moraxella catarrhalis* [9, 10]. In patients with bronchiectasis, *Haemophilus influenzae* and *P. aeruginosa* are reported to more commonly co-exist with *Candida albicans* and *Aspergillus fumigatus.* The role of fungal infections in bronchiectasis patients with bacterial colonization remains insufficiently examined.

The prognostic impact of putative invasive pulmonary aspergillosis (IPA) in COPD patients was previously investigated in a study comparing 50 IPA cases with non-IPA cases. The results indicated poorer survival outcomes in IPA patients, while bacterial etiologic agents did not demonstrate a significant prognostic effect [11]. Amidst the increasing prevalence of CPA, there is limited data on the bacterial spectrum in CPA patients, particularly in terms of chronic colonization and co-infection. Furthermore, the incidence and prognostic impact of bacterial co-infections in CPA patients have yet to be clearly established.

The objective of this study is to investigate the incidence and associated factors of bacterial co-infection in CPA patients, as well as its potential prognostic impact on this population.

Materials and methods

In this retrospective, single-center, observational study, individuals with an ICD-10 code of B44.18 (pulmonary aspergillosis) diagnosed between January 2019 and January 2024, were reviewed (Fig. 1).

Diagnostic criteria

CPA diagnosis was established based on the criteria outlined in current guidelines (42,43). Only CPA patients who had undergone bacterial culture testing within seven days of diagnosis were included.

Co-infections were defined as infections occurring within seven days before or after CPA diagnosis. A co-infection was determined based on the presence of clinical and/or radiological signs of infection and the identification of a relevant pathogen of significant amount within this timeframe [12].

Organization of the clinics

The hospital where this study was conducted is a reference center for chest diseases and has the largest bed capacity in the country. The inpatient and outpatient clinics provide continuous healthcare services to both local patients and referrals from across the country.

Regarding CPA, the diagnosis, treatment, and follow-up plans of all patients are evaluated by a multidisciplinary council consisting of specialists in chest diseases, infectious diseases, and thoracic surgery. Additionally, when necessary or upon request, pathologists and microbiologists also participate in these meetings.

In this study, only patients with microbiologically analyzed sputum and/or bronchoscopic samples or tracheal aspirates, assessed using Gram staining and bacterial culture, were included. In our hospital, bronchoscopic sampling is performed from the lesion site identified on radiological imaging using bronchoscopic aspirate or bronchoalveolar lavage. All samples are delivered to the laboratory as soon as possible after collection. The cellular quality of the samples and bacterial counts in deep respiratory tract specimens are evaluated according to the guidelines. When a microorganism is identified as a pathogen, treatment is administered based on antibiotic susceptibility test results [13].

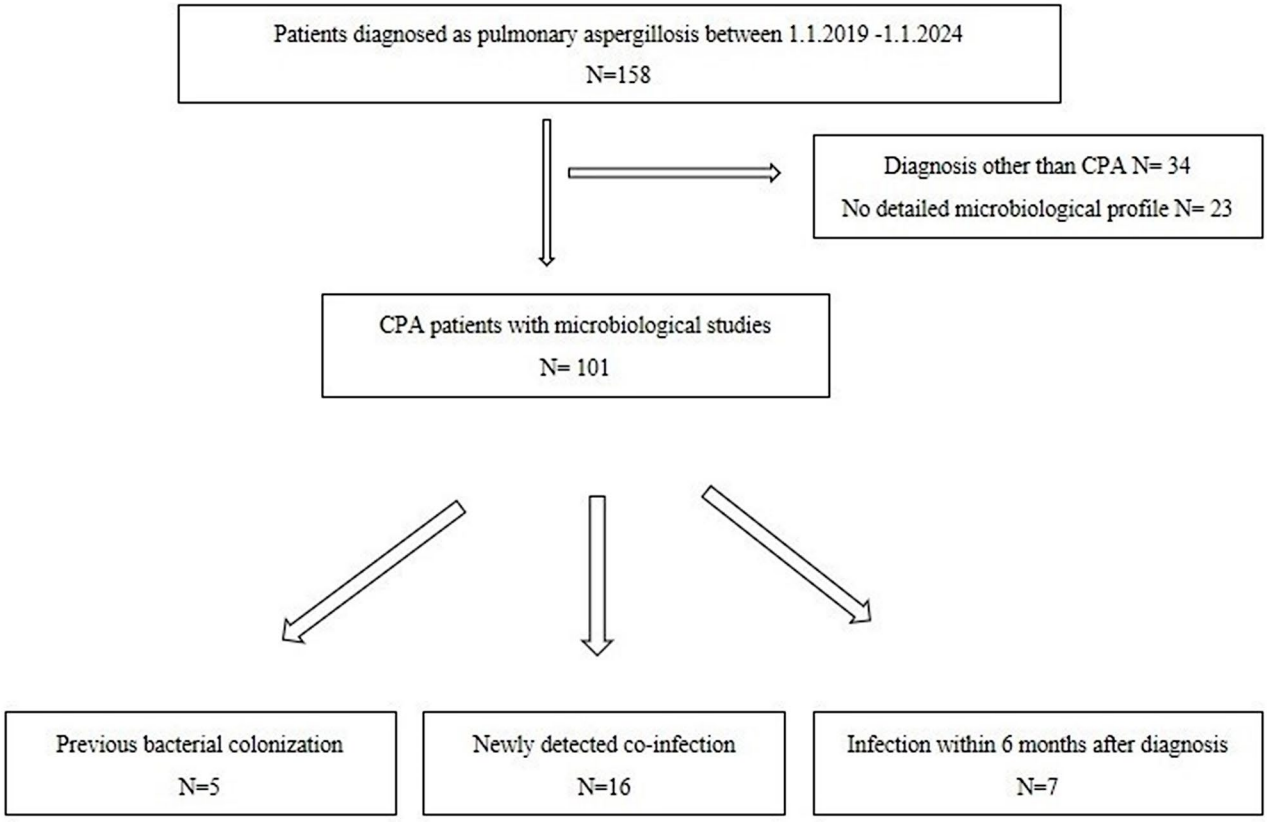

Fig. 1 Flowchart of the patients

Data collection and study design

Demographic data, additional diseases, smoking status and smoking load of the patients were recorded. Presenting symptoms, including the presence and amount of hemoptysis, were noted. Radiological findings, including pleural effusion, laterality of pulmonary involvement (right, left or bilateral) and lobar involvement (right upper lobe [RUL], right lower lobe [RLL], left upper lobe [LUL], left lower lobe [LLL], or multilobar), were analyzed. Bronchoscopic procedures were undertaken and findings were recorded. Diagnostic method and pathological diagnostic procedures were registered.

Based on radiological and clinical findings, patients were classified into the following CPA subtypes: aspergilloma, chronic cavitary pulmonary aspergillosis (CCPA), chronic fibrosing pulmonary aspergillosis (CFPA), subacute invasive pulmonary aspergillosis (SAIA), and aspergillosis nodule [14, 15]. Tracheobronchial aspergillosis was also documented.

Galactomannan levels at diagnosis, treatment modalities, and antifungal therapy details were recorded. Patient readmissions and survival status during follow-up were documented.

To identify factors associated with bacterial co-infections, patients were categorized into co-infected and non-co-infected groups. General characteristics and recorded parameters of the two groups were compared. Variables found to be significant in univariate analysis were subsequently included in multivariate analysis to determine independent predictors of co-infection.

Definitions

Bacterial co-infection: The presence of any microbiological growth within seven days before or after the diagnosis of pulmonary aspergillosis, which was accepted as a pathogen.

Chronic colonization: The identification of the same bacterial species in sputum and/or bronchoscopic cultures on at least two occasions within one year. In this study, five patients were classified as having chronic colonization. However, since these patients were also considered infectious at the time of presentation, they were categorized under the co-infection group.

Infection within 6 months: Any newly detected bacterial growth occurring between the second week and the sixth month following the diagnosis of pulmonary aspergillosis. In this group, three patients with post-operative bacterial detection were excluded from the analysis.

Table 1 Comparison of CPA patients with and without bacterial co-infection

	All CPA patients N = 101	Bacterial Co-infection (N = 21)	No Bacterial Co-infection (N = 80)	P
	N (%)	N (%)	N (%)	
Male gender	79	15 (72)	64 (80)	0.388
Age (year)	57 ± 13 (28–80)	57 ± 12	57 ± 16	0.939
Ever smoking rate	69	16 (80)	53 (75)	0.772
Smoking load (pack/year)	37 ± 20 (5-110)	35 ± 16	45 ± 30	0.087
Symptoms at presentation				
Dyspnea	56	15 (71)	41 (51)	0.139
Cough	57	14 (67)	43 (54)	0.331
Hemoptysis	56	10 (48)	46 (58)	0.466
Sputum	17	8 (38)	9 (11)	**0.007**
Weight loss	7	3 (14)	4 (5)	0.155
Amount of hemoptysis				
Bloody-streaked	33	5 (24)	28 (35)	
Tea-spoon or table-spoon	15	3 (14)	12 (15)	0.809
Cup	8	2 (10)	6 (8)	
Respiratory failure	7	5 (24)	2 (3)	**0.007**
Any additional diseases	90	18 (86)	72 (90)	0.694
ICU admission	4	2 (10)	2 (3)	0.19
Systemic steroid use	10	5 (24)	5 (6)	**0.03**
COPD	33	6 (29)	27 (34)	0.796
CAD	5	1 (5)	4 (5)	NS
Asthma	3	-	3 (4)	NS
Bronchiectasis	8	3 (14)	5 (6)	0.358
DM	24	4 (19)	20 (25)	0.755
Malignancy	17	3 (14)	14 (18)	NS
Emphysema	11	4 (19)	7 (9)	0.233
TB Sequela	46	6 (29)	40 (50)	0.091
ILD	8	3 (14)	18 (86)	0.358
CTD	5	1 (5)	4 (5)	NS
Any organ transplant	2	-	2	NS
Pleural effusion	7	1 (5)	6 (8)	NS
Side of involvement				
Right	45	10 (48)	35 (44)	
Left	37	7 (33)	30 (38)	0.934
Bilateral	19	4 (19)	15 (19)	
Lobar involvement				
RUL	37	8 (38)	29 (36)	
LUL	31	4 (19)	27 (34)	0.162
LLL	3	2 (10)	1 (1)	
Multiple lobes	30	7 (33)	23 (29)	
TTNAB	6	0	6 (8%)	0.34
Bronchoscopy	75	17 (81)	58 (72)	0.054
Daignosis method				
Laboratory	14	3 (14)	11 (14)	
Radiology and clinical	14	7 (33)	7 (9)	0.013
Pathology	73	11 (53)	62 (77)	
Pathological diagnosis via				
TTNA	4		19 (24)	
Surgical	47	4 (19)	4 (5)	**0.017**
Bronchoscopic biopsy	6	1 (5)	43 (54)	
Bronchocopic lavage	15	6 (29)	9 (11)	
Radiological classification				
Simple aspergilloma	43	7 (33)	36 (45)	

Table 1 (continued)

	All CPA patients N = 101	Bacterial Co-infection (N = 21)	No Bacterial Co-infection (N = 80)	P
	N (%)	N (%)	N (%)	
CCPA	26	5 (24)	21 (26)	
CFPA	5	3 (14)	2 (3)	0.198
SAIA	9	2 (10)	7 (9)	
Aspergillosis nodule	8	3 (14)	5 (6)	
Endobronchial aspergillosis	10	1 (5)	9 (11)	
Galactomannan serum	56	10	46	0.715
High levels (> 1)	13	3	10	
Galactomannan level	0.867 ± 1.36 (0.015–6.18)	0.949 ± 1.47	0.849 ± 1.35	0.835
Galactomannan BAL	18	3	15	0.685
High (> 1)	17	3	14	
Galactomannan level (BAL)	4.926 ± 4.63 (0.431–15.225)	2.8 ± 0.5	5.3 ± 4.9	0.401
Treatment Method				
Follow-up	8	3 (14)	5 (6)	
Surgery alone	34	4 (19)	30 (38)	0.047
Antifungal Treatment alone	42	13 (62)	29 (36)	
Treatment and surgery	17	1 (5)	16 (20)	
Antifungal treatment				
Fluconazole	1		1	
Voriconazole	50	13 (62)	37	0.864
Capofungin	1		1	
Itraconazol	7	1 (5)	6	
Antifungal treatment complication	9	2 (10)	7 (16)	NS
Surgical operation	51	5	46	0.357
Number of hospitalisation				
1–2	93	19 (90)	74 (93)	0.67
3 and more	8	2 (10)	6 (7)	
Follow-up duration (months)	27 ± 22 (0–83)	25.7 ± 20.6	28.8 ± 25	0.564
Survival duration (months)	37 ± 25 (0–83)	35.1 ± 27	37.2 ± 23	0.736
Mortality	29	9 (43%)	20 (25%)	0.174
Malignancy	9			

BAL: bronchoalveolar lavage, CAD: coronary artery disease, CCPA: chronic cavitar pulmonary aspergillosis, CFPA: chronic fibosing pulmonary aspergillosis, COPD: chonic obstructive pulmonary disease, CTD: connective tissue disease, DM: diabetes mellitus, ICU: intensive care unit, ILD: interstitial lung disease, LLL: left lower lobe, LUL: left upper lobe, RUL: right upper lobe, SAIA: subacute invasive aspergillosis, TB: tuberculosis, TTNAB: transthoracic needle aspiration biopsy

Statistical analysis

Quantitative data are expressed as mean ± standard deviation (SD) and qualitative data are expressed as frequencies. Student's t-test and chi-square tests were used to assess potential factors associated with bacterial co-infection.

ll-cause mortality was evaluated for both in-hospital and long-term outcomes. Survival curves were drawn using the Kaplan–Meier method. Cox's proportional hazards model was used to determine potential predictors of mortality. Independent variables associated with respiratory mortality with a P value < 0.05 in the univariate analysis were then incorporated into a multivariate analysis, also based on the Cox proportional hazards model.

All statistical analyses were conducted using a statistical software package (SPSS for Windows, version 16.0; SPSS Inc.; Chicago, IL, USA). A P value of < 0.05 was considered statistically significant.

Results

Of all the 101 patients, the mean age was 57 ± 13 years (range: 28–80) and 79 were male. The rate of ever cigarette smoking was 69%, and 90% of the patients had at least one additional comorbidity. At presentation, 56 patients reported hemoptysis (Table 1).

Co-infection rate was recorded in 21 patients (21%). The samples were analyzed using bronchoscopic aspirate in 6 (28%) patients and sputum cultures in 13 patients (62%), while both sputum and bronchoscopic aspirate yielded the same bacteria in 2 patients (10%). The most frequently isolated bacteria at diagnosis were *P. aeruginosa* ($n = 6$), *K. pneumoniae* ($n = 5$), *Escherichia coli* ($n = 4$), *and Serratia marcescens* ($n = 4$). Five patients

Table 2 Microbiological profile of patients with co-infection at diagnosis and follow-up

	Chronic colonization ($n=5$)*	Newly detected co-infection ($n=16$)**	Infection within 6 months after diagnosis ($n=7$)
Pseudomonas aeruginosa	3	3	2
Klebsiella pneumoniae	2	3	5****
Escherichia coli		4	1****
Serratia marcescens	2	2	
Stenetrophomonas maltophilia		2	1
Enterobacter spp.		1***	
Acinetobacter baumanii		1	1
Staphylococcus aureus		1	
Enterococcus faecalis		1	
Morganella morganii			1

*: One case was chronically colonised with *P. aeruginosa* and *K. pneumoniae*, while other one patient was colonised with *K. pneumoniae* and *S. marcescens*

**: One case: Co-infected with *Stenetrophomonas maltophilia* and *E. Coli*, One case: *Enterobacter spp.* and *K. pneumoniae*, One case: *K. pneumoniae* and *Acinetobacter baumanii*

***: This case was also co-infected with *Mycobacterium Tuberculosis*

****: Two patients were diagnosed in the post-operative period

were previously colonised with bacteria of: *P. aeruginosa* ($n=1$), *Serratia marcescens* ($n=2$) and *K. pneumoniae* ($n=3$). At diagnosis, more than one bacterial species were detected in six patients. After diagnosis, five *K. pneumoniae* cases and two cases of *P. aeruginosa* infections were identified. (Table 2). Patients' antibiotic treatments included piperacillin-tazobactam ($n=8$, combined with fluoroquinolone in 3 patients), meropenem ($n=7$, combined with polymyxin B in 2 patients and with fluoroquinolone in 1 patient), ceftriaxone ($n=3$, with fluoroquinolone in 2 patients), and ceftazidime ($n=2$, with fluoroquinolone in 1 patient). The mean duration of intravenous antibiotic treatment was 15.4 ± 5 days (range: 8–26 days).

A comparison between bacterial co-infected and non co-infected patients revealed similar demographics and smoking status. At presentation, bacterial co-infected patients had more frequent sputum complaints (34% vs. 11%, $p=0.007$) and hypoxemic respiratory failure (24% vs. 3%, $p=0.007$). The presence and amount of hemoptysis did not differ between the groups ($p>0.05$). Systemic corticosteroid use was more common in the co-infected group (24% vs. 6%, $p=0.03$) at presentation. Radiological localization and the presence of pleural effusion were not associated with bacterial co-infection ($p>0.05$).

Seventeen patients with bacterial co-infection had undergone bronchoscopy, and 11 were diagnosed based on pathological findings. Diagnostic procedures and methods did not significantly differ between the groups. Radiologically, in the co-infected group, 7 patients (33%) had aspergilloma, 5 (24%) had chronic cavitary pulmonary aspergillosis (CCPA), and 2 had SAIA.

In terms of treatment, sole antifungal treatment was more frequent in the co-infected group, while surgical operations were more commonly performed in the non-co-infected group (62% vs. 36%, respectively, $p=0.047$). After diagnosis, overall follow-up interval was 27 ± 22 months (range: 2–83). The co-infected group was followed for 25.7 ± 18.6 months, and the non co-infected group for 28.8 ± 22 months. With similar follow-up durations, the number of hospitalizations, mortality, and survival time did not differ significantly between the two groups ($p>0.05$). The mean survival time was 55.8 ± 8.2 in co-infected patients and 63.3 ± 3.5 in non-co-infected patients (HR: 0.623 CI: 0.272–1.424, $p=0.254$) (Fig. 2).

When including patients with bacterial co-infection within 6 months, the mean survival time was 64.5 ± 3.6 months in the non-co-infected group and 53.8 ± 7 months in the co-infected group (Log-rank test: 0.118) (Fig. 3). Previously colonized patients experienced shorter survival than non-colonized patients; however, this difference was not statistically significant (64.1 ± 3.5 vs. 44.4 ± 12.5 months, respectively, Log-rank test: 0.071) (Fig. 4).

Discussion

The present study is the first of its kind to examine the microbiological profile, particularly in CPA patients, focusing on potential associated factors and the prognostic impact of bacterial co-infection. Among 101 patients, five had previously documented bacterial colonization, most frequently with *P. aeruginosa.* In 16 pateints, new co-infections were recorded, most commonly caused by *Escherichia coli*, *K. pneumoniae* and *P. aeruginosa.* During a six-month follow-up period, new co-infections were observed in seven patients, predominantly with *K. pneumoniae.* No significant association was observed between bacterial infections and increased mortality in CPA patients.

According to one very recent study, a middle-aged woman with COPD and diabetes mellitus was reported to have a co-infection. At presentation, she exhibited symptoms of infection and had acute type 2 respiratory failure. Co-infection with *P. aeruginosa* and *Aspergillus fumigatus* was confirmed, and she was treated with both antibacterial and antifungal therapy, resulting in a full recovery [16]. Rawson et al. reviewed COVID-19 related infections and reported a bacterial and fungal co-infection rate of 8% [5]. Another study focused on 86 possible cases of COVID-19 related pulmonary aspergillosis (CAPA) and noted that 47% of these patients may have had bacterial co-infections. The most common bacteria identified were *Enterobacteriaceae* (55%, $n=22/40$), followed by non-fermenting Gram-negative rods (30%,

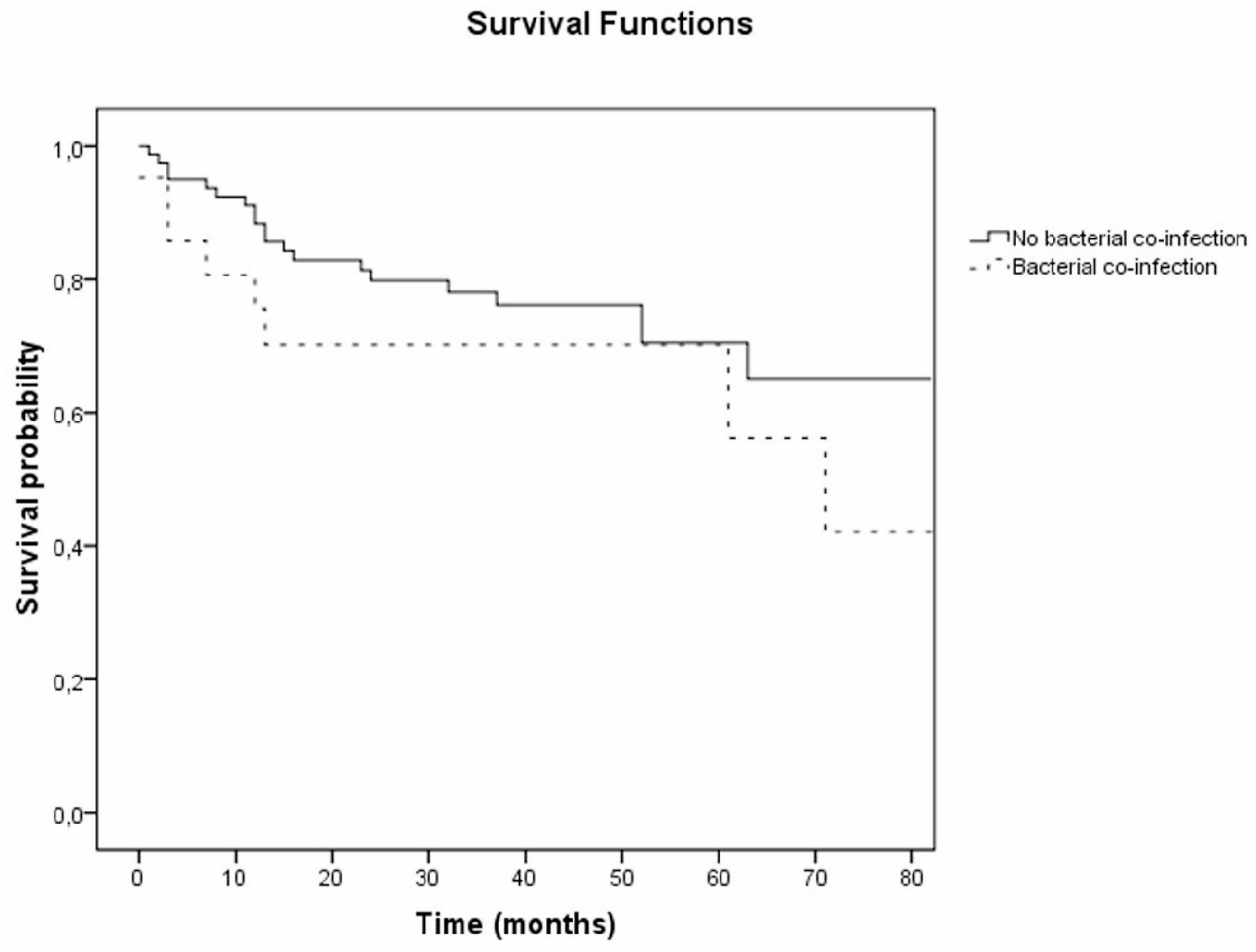

Fig. 2 Survival curves of patients with and without bacterial co-infection

$n = 12/40$). The most frequent species responsible for these co-infections were *Escherichia coli*, *P. aeruginosa*, *Acinetobacter baumannii*, and *K. pneumoniae* [17]. These reports underline the fact that bacterial and fungal infections are not uncommon.

In our study, more than one in five pulmonary aspergillosis patients were also co-infected with bacteria. The higher rate in the aforementioned study may be attributed to the specific group of COVID-19 patients, as viral pneumonia has been shown to exacerbate the risk of bacterial and fungal co-infections. Still, given the retrospective nature of our study, it is likely that not all respiratory tract samples for co-infections were collected before antibiotic use. Therefore, the co-infection rates may have been underestimated. Nevertheless, the frequency of bacterial co-infections with pulmonary aspergillosis remains subject to further investigation. It can be hypothesized that early detection of both infections and appropriate treatment may prevent further complications.

In our study, the most frequently detected bacterial co-infections were due to *Escherichia coli*, *K. pneumoniae*, and *P. aeruginosa*. Interestingly, during follow-up, *K. pneumoniae* infection was common. No existing literature could be found, to the best of our knowledge, to shed light on this issue. However, a noteworthy case was recently reported: A 66-year-old man with COPD presented with severe pneumonia and cardiac arrest and was diagnosed with both *K. pneumoniae* and *Aspergillus fumigatus* infection. This report is the first one documenting this type of co-infection [8]. We believe further studies are needed on the potential relationship of the bacterial elements in this co-infection.

In the literature, polymicrobial infections are more frequently studied in specific patient groups. In cystic fibrosis (CF), bacterial infections are defined as a risk factor for developing fungal infections [6]. The potential correlation between putative IPA and previous bacterial infections was investigated in intensive care unit (ICU) patients, considering both previous colonization and infections within 6 months prior to ICU admission. No significant difference in bacterial colonization or infection was found between patients and control groups. In

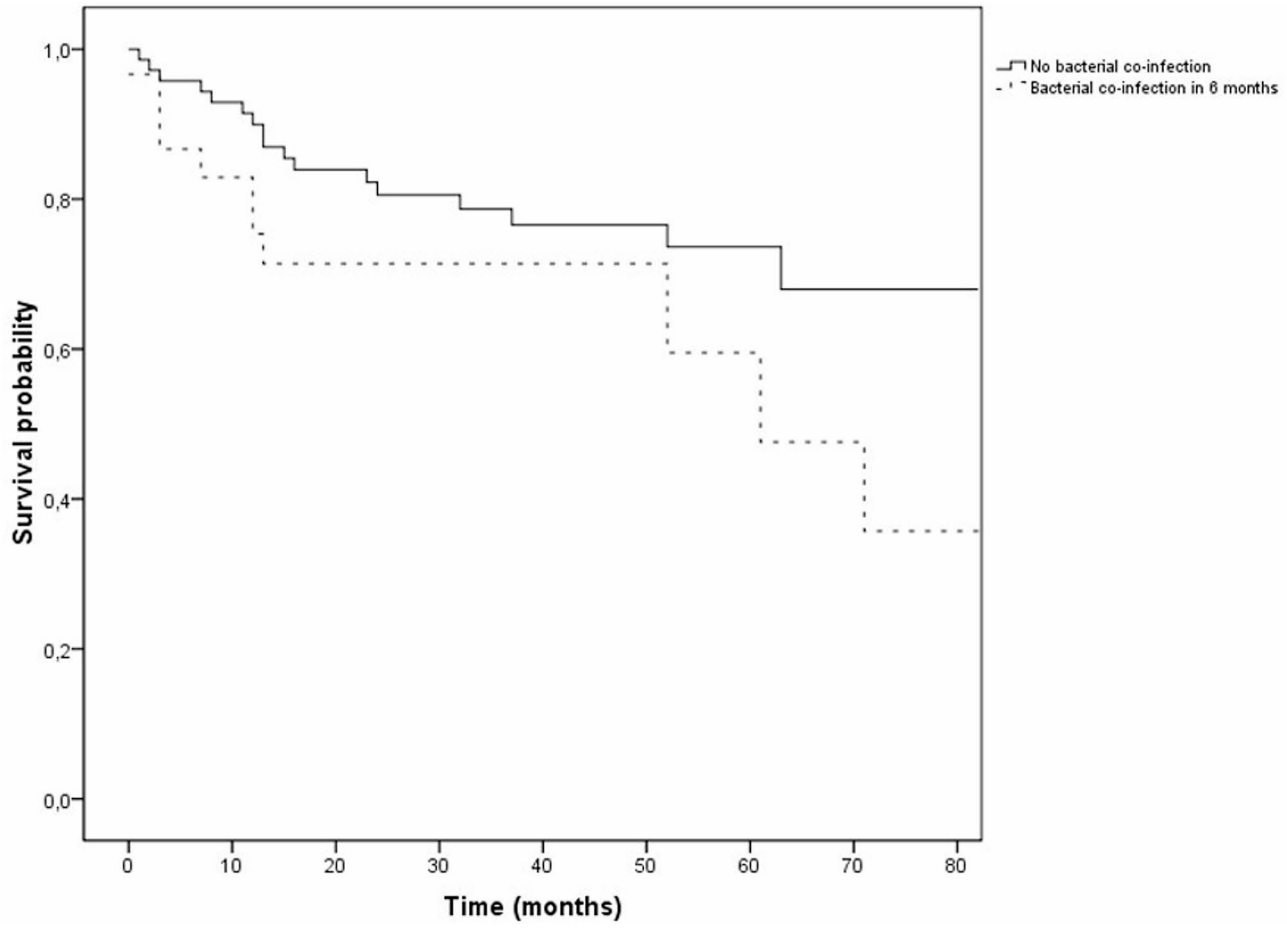

Fig. 3 Survival curves of patients according to bacterial co-infections within 6 months after diagnosis ($p=0.118$)

IPA group, 60% had any bacterial growth evaluated as colonization or infection. The most frequent pathogens included non-fermenting bacteria (40%), primarily *Pseudomonas spp.* (28%) along with *Acinetobacter spp.*, *Stenotrophomonas spp.*, *Burkholderia spp. enterobacteria*, and gram positive bacteria (20%) including *Staphylococcus aureus*, *Streptococcus pneumoniae*, and *Enterococcus spp.*) [11].

Iqbal N, et al. studied microbiological profile of ABPA patients. Among 245 ABPA patients, sputum cultures were examined in 103, with the most common pathogens identified as *P. aeruginosa* (31.1%) and *Haemophilus influenzae* (15.5%) [18]. It is known that ABPA can alter to CPA [19], making the findings of this study, which suggest a possible ABPA-bacterial co-infection, particularly relevant for CPA patients.

Danion F. et al., investigated bacterial co-infection in invasive aspergillosis, finding a co-infection rate of 16% among 690 patients. The co-infection site was pulmonary in 48% of cases. Bacteria in these co-infections included *Staphylococcus aureus* (4%), coagulase-negative *Staphylococci* (7%), *Streptococcus* and *Enterococcus* (7%), *Escherichia coli* (7%), *P. aeruginosa* (6%), other *Enterobacteria* (5%), *K. pneumoniae* (3%), other non-fermenting gram-negative bacilli (2%), other bacteria 13 (3%), and *Mycobacteria* 6 (2%) [12]. Autopsy studies of hematological malignancy (HM) patients have identified bacterial and fungal co-infections in 42–46% of cases [20, 21]. In our study, only CPA patients were included, with a focus on pulmonary bacterial co-infections. The observed differences in rates and microbiological profiles of the patients may be attributed to variations in the patient populations.

Possible asssociated factors with co-infection were studied in IPA. In a multivariate analysis; allogeneic haematopoietic stem-cell transplantation (alloHSCT), other HM, the presence of other underlying diseases, non-nodular lesions on CT, low lymphocyte count, high C-reactive protein levels, fever, tracheal intubation, and isolation of two or more different species of *Aspergillus* were correlated with co-infections. In contrast, age > 65 years was associated with a lower rate of co-infection [12]. Another study focusing on co-infections in HM patients did not report any differences in terms of subpopulation and radiological characteristics between patients with or without co-infections [22]. In our study, only CPA patients were included. Sputum production,

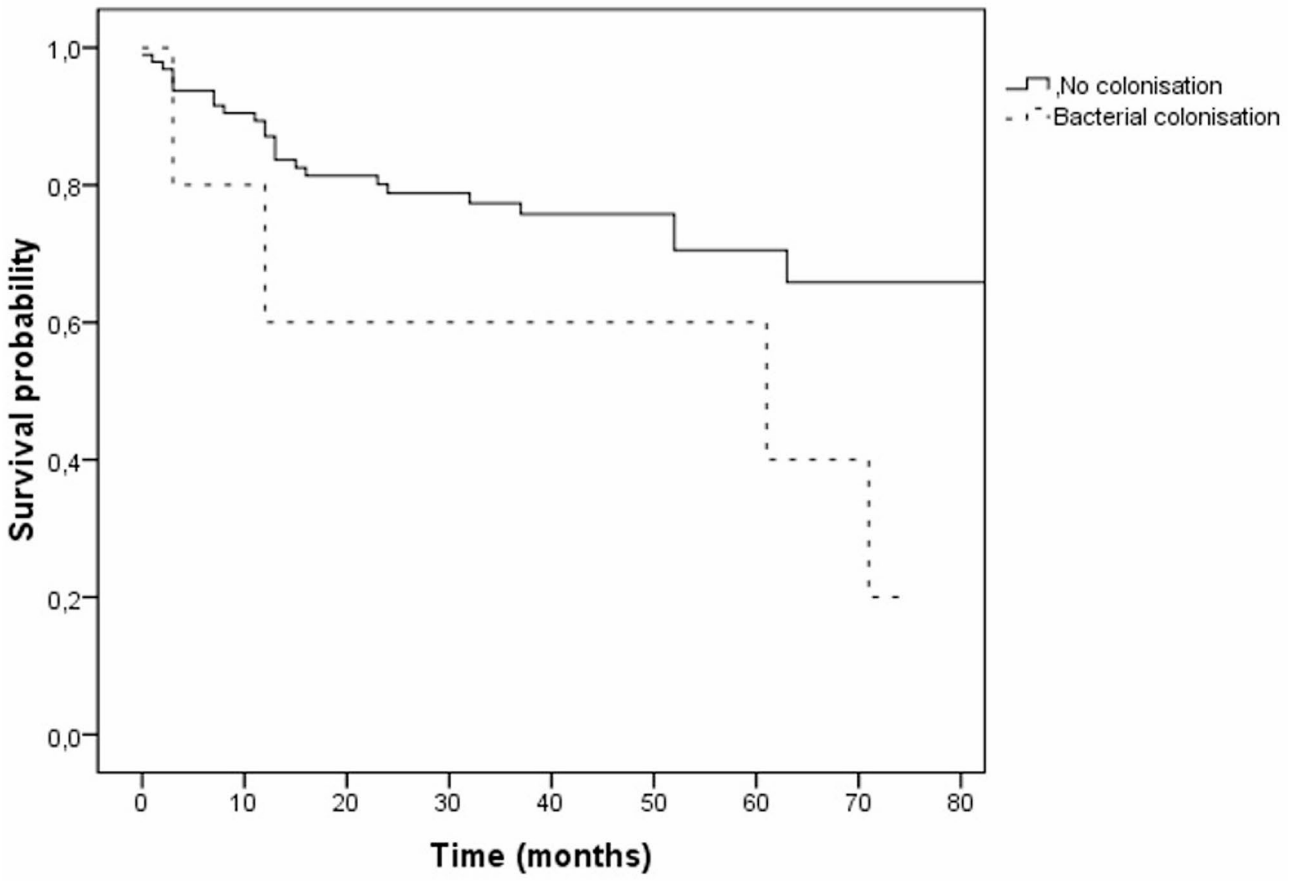

Fig. 4 Survival curves of patients with and without previously bacterial colonisation (64.1 ± 3.5 vs. 44.4 ± 12.5, respectively, Log-rank test: 0.071)

hemoptysis at presentation, and systemic steroid use were more common in bacterial co-infections. While laboratory values were not studied for the purposes of ourstudy, further research may reveal the impact of various blood parameters in CPA patients.

The underlying etiology of the possible relationship between co-infection is yet to be fully understood. Bargon et al. demonstrated a significant relationship between inhaled/oral antibiotics and *Aspergillus* colonization [23]. They attributed this association to the antagonistic relationship between *P. aeruginosa* and *Aspergillus* spp. It has been reported that *P. aeruginosa* species inhibit *A. Fumigatus* filamentation by releasing extracellular molecules that disrupt intracellular communication, thus limiting fungal growth [24]. A case report published suggests that co-infection with CPA and *P. aeruginosa* may lead to angioinvasion and progressive and recurrent pneumonia [25]. In contrast, an Irish CF study found a 3.1% co-colonisation rate of *Aspergillus fumigatus* and *P. aeruginosa*. This co-infection resulted in higher exacerbations, hospitalisations and antimicrobial usage compared to patients solely colonised with *Pseudomonas spp*. However, the prognosis of solely *Aspergillus*-colonized patients did not show poorer outcomes [7]. An imbalance between pro-inflammatory and anti-inflammatory cytokines may occur during infections. Unfortunately, the method used in our own study does not allow conclusions regarding *Aspergillus* colonization and *P. aeruginosa* infections. Nevertheless, clarifying the pathogenic relationship in patients with fungal infections is essential, as specific bacteria play a prominent role in the infections of CPA patients.

Studies on the prognosis of the co-infections are rather limited in number. In patients with HM, co-infections were found to be independently associated with increased overall and hospital mortality [26, 27]. In terms of survival in invasive aspergillosis patients, bacterial co-infected patients are reported to have lower 12-week mortality in univariate analysis (HR 1.4, 95%CI 1.02–1.9). However, in multivariable analysis, co-infection was independently identified as a risk factor for 12-week mortality (adjusted HR 1.5, 95% CI 1.1–1.9, $p < 0.01$) [12]. Studies with conflicting results regarding prognosis have also been reported. Putative IPA in COPD patients, when compared with cases without IPA, demonstrated worse overall survival. However, bacterial etiologic agents did not yield any impact on the prognosis [11].

With a different study method and distinct patient groups compared to aforementioned studies, our study did not find a correlation between bacterial co-infections and mortality in CPA patients. One reasonable explanation for such a lack of correlation could be that timely diagnosis, appropriate treatment choices, and close monitoring prevented a negative prognosis. While the general expectation for co-infected patients is a more severe disease course, one could hypothesize that bacterial toxins may potentially dampen or decelerate the progression of Aspergillosis, thus preventing a worsened prognosis. We believe that such questions are open to debate and warrant further in-depth research. Multi-center studies with larger patient populations are needed, firstly, to focus on the prognostic effect of co-infections, and secondly, to test these potential hypotheses.

There are some limitations to our study. Firstly, it is a retrospective, single-center study. Secondly, conducting a detailed search for viral infections was not feasible. The performance of viral infection tests is strictly regulated in our country. The third limitation is that prior antibiotic treatment and antibiotic resistance profiles were not recorded. However, all patients were included in the study and treated based on clinically, laboratory, and microbiologically confirmed pathogens. The final limitation is; considering the likelihood of prior antibiotic use, some co-infections may have been undetected, potentially leading to an underestimation of the co-infection rate. On the other hand, the main strength of our study is that it was conducted in a reference chest disease center. The demographics and socio-economic diversity of the patient population make our findings more reflective of real-life conditions, thereby enhancing the robustness of our research. Moreover, in our center, patients are managed in an integrated fashion and are continuously under close scrutiny, in line with the Professional Council guidelines. Finally, our study contributes to the literature with its large patient cohort, promising methodology, and robust design.

In conclusion, this study, conducted on a large group of CPA patients, finds that more than one in five patients with CPA may have bacterial co-infections. Still, the prognosis does not appear to differ significantly when compared to non-co-infected patients. Early diagnosis and appropriate treatment for both pathogens, along with close management and follow-up, may contribute to similar prognoses in these patients.

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Author contributions
Conceptualization: F.TA., D.H.G. Methodology: F.T.A, H.A, SS. Data collector: S.G., H.A., Ç.S. Statistical Analysis: F.T.A, S.S Writing original draft: F.T.A, S.A. Supervision: H.A., D.H.G, S.S., S.A. Writing review and editing: F.T.A., S.G., H.A., D.H.G, Ç.S., S.S., S.A. All authors reviewed the manuscript.

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Data availability
Data are available from the corresponding author (F.T.A) upon reasonable request.

Declarations

Ethics approval and consent to participate
The study protocol was approved by the Yedikule Chest Diseases and Thoracic Surgery Training and Research Hospital Scientific and Ethics Committe (8.11.2023, 359-2) and performed under the Declaration of Helsinki. Due to the retrrospective nature of the study, the obligation to obtain informed consent was waived by the Ethics Committee.

Competing interests
The authors declare no competing interests.

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References

1. Denning DW. Global incidence and mortality of severe fungal disease. Lancet Infect Dis. 2024;24:e269.
2. Tashiro M, Takazono T, Izumikawa K. Chronic pulmonary aspergillosis: comprehensive insights into epidemiology, treatment, and unresolved challenges. Ther Adv Infect Dis. 2024;11:20499361241253751.
3. Amin R, Dupuis A, Aaron SD, Ratjen F. The effect of chronic infection with Aspergillus fumigatus on lung function and hospitalization in patients with cystic fibrosis. Chest. 2010;137(1):171–6.
4. Lee TWR, Brownlee KG, Conway SP, Denton M, Littlewood JM. Evaluation of a new definition for chronic Pseudomonas aeruginosa infection in cystic fibrosis patients. J Cyst Fibros. 2003;2(1):29–34.
5. Rawson TM, Moore LSP, Zhu N, Ranganathan N, Skolimowska K, Gilchrist M, Satta G, Cooke G, Holmes A. Bacterial and fungal coinfection in individuals with coronavirus: A rapid review to support COVID-19 antimicrobial prescribing. Clin Infect Dis. 2020;71(9):2459–68.
6. Magee LC, Louis M, Khan V, Micalo L, Chaudary N. Managing fungal infections in cystic fibrosis patients: challenges in clinical practice. Infect Drug Resist. 2021;14:1141–53. https://doi.org/10.2147/IDR.S267219.
7. Reece E, Segurado R, Jackson A, McClean S, Renwick J, Greally P. Co-colonisation with Aspergillus fumigatus and Pseudomonas aeruginosa is associated with poorer health in cystic fibrosis patients: an Irish registry analysis. BMC Pulm Med. 2017;17(1):70.
8. Bhatia IPS, Singh A, Hasvi J, Rajan A, Venigalla SK. Coinfection of Klebsiella pneumoniae and Aspergillus in a patient with chronic obstructive pulmonary disease post cardiac arrest: a case report. J Med Case Rep. 2024;18(1):427.
9. Máiz L, Nieto R, Cantón R, Gómez G, de la Pedrosa E, Martinez-García MÁ. Fungi in bronchiectasis: A concise review. Int J Mol Sci. 2018;19(1):142.
10. McDonnell MJ, Jary HR, Perry A, MacFarlane JG, Hester KL, Small T, Molyneux C, Perry JD, Walton KE, De Soyza A. Non cystic fibrosis bronchiectasis: A longitudinal retrospective observational cohort study of Pseudomonas persistence and resistance. Respir Med. 2015;109:716–26.
11. Delsuc C, Cottereau A, Frealle E, Bienvenu AL, Dessein R, Jarraud S, Dumitrescu O, Le Maréchal M, Wallet F, Friggeri A, Argaud L, Rimmelé T, Nseir S, Ader F. Putative invasive pulmonary aspergillosis in critically ill patients with chronic obstructive pulmonary disease: a matched cohort study. Crit Care. 2015;3(19):421.
12. Danion F, Duval C, Séverac F, Bachellier P, Candolfi E, Castelain V, Clere-Jehl R, Denis J, Dillenseger L, Epailly E, Gantzer J, Guffroy B, Hansmann Y, Herbrecht JE, Letscher-Bru V, Leyendecker P, Le Van Quyen P, Ludes PO, Morel G, Moulin B, Paillard C, Renaud-Picard B, Roche AC, Sabou M, Schneider F, Solis M, Talagrand-Reboul E, Veillon F, Ledoux MP, Simand C, Herbrecht R. Collaborators. Factors associated with coinfections in invasive aspergillosis: a retrospective cohort study. Clin Microbiol Infect. 2021;27(11):1644–51.
13. Baquero F, Alvarez ME, Cantón R. Bacteriologic diagnosis of respiratory tract infection. Clin Microbiol Infect. 1996;1:S210–5.

14. Denning DW, Cadranel J, Beigelman-Aubry C, Ader F, Chakrabarti A, Blot S, Ullmann AJ, Dimopoulos G, Lange C. European society for clinical microbiology and infectious diseases and European respiratory society. Chronic pulmonary aspergillosis: rationale and clinical guidelines for diagnosis and management. Eur Respir J. 2016;47(1):45–68.
15. Barac A, Vujovic A, Drazic A, Stevanovic G, Paglietti B, Lukic K, Stojanovic M, Stjepanovic M. Diagnosis of chronic pulmonary aspergillosis: clinical, radiological or laboratory?? J Fungi (Basel). 2023;9(11):1084.
16. Das D, Panda PK. Pseudomonas and Aspergillus symbiotic coinfections in a case of chronic obstructive pulmonary disease and diabetes mellitus. BMJ Case Rep. 2024;1(7):e259285.
17. Hernández-Silva G, Corzo-León DE, Becerril-Vargas E, Peralta-Prado AB, Odalis RG, Morales-Villarreal F, Ríos-Ayala MA, Alonso TG, Agustín FD, Ramón AF, Hugo AV. Clinical characteristics, bacterial coinfections and outcomes in COVID-19-associated pulmonary aspergillosis in a third-level Mexican hospital during the COVID-19 pre-vaccination era. Mycoses. 2024;67(1):e13693.
18. Iqbal N, Irfan M, Bin Ali Zubairi M et al. Allergic bronchopulmonary aspergillosis: radiological and Microbiological profile of patients presented in an outpatient pulmonary clinic in a developing country. Monaldi Arch Chest Dis. https://doi.org/10.4081/monaldi.2023.2803
19. Denning DW, Pleuvry A, Cole DC. Global burden of allergic bronchopulmonary aspergillosis with asthma and its complication chronic pulmonary aspergillosis in adults. Med Mycol. 2013;51:361–70.
20. Chamilos G, Luna M, Lewis RE, Bodey GP, Chemaly R, Tarrand JJ, et al. Invasive fungal infections in patients with hematologic malignancies in a tertiary care cancer center: an autopsy study over a 15-year period (1989e2003). Hae Matologica. 2006;91:986e9.
21. Lewis RE, Cahyame-Zuniga L, Leventakos K, Chamilos G, Ben-Ami R, Tamboli P, et al. Epidemiology and sites of involvement of invasive fungal infections in patients with haematological malignancies: a 20-year autopsy study. Mycoses. 2013;56:638e45.
22. Georgiadou SP, Kontoyiannis DP. Concurrent lung infections in patients with hematological malignancies and invasive pulmonary aspergillosis: how firm is the Aspergillus diagnosis? J Infect. 2012;65:262e8.
23. Bargon J, Dauletbaev N, Köhler B, Wolf M, Posselt HG, Wagner TO. Prophylactic antibiotic therapy is associated with an increased prevalence of Aspergillus colonization in adult cystic fibrosis patients. Respir Med. 1999;93(11):835–8.
24. Mowat E, Rajendran R, Williams C, et al. Pseudomonas aeruginosa and their small diffusible extracellular molecules inhibit Aspergillus fumigatus biofilm formation. FEMS Microbiol Lett. 2010;313(2):96–102.
25. Mahendran M, Urbine D. Invasive pulmonary aspergillosis/pseudomonas. BMJ Case Rep. 2021;14(7):e236887.
26. Hardak E, Avivi I, Berkun L, Raz-Pasteur A, Lavi N, Geffen Y, et al. Polymicrobial pulmonary infection in patients with hematological malignancies: prevalence, co-pathogens, course and outcome. Infection. 2016;44:491e7.
27. Burghi G, Lemiale V, Seguin A, Lambert J, Lacroix C, Canet E, et al. Outcomes of mechanically ventilated hematology patients with invasive pulmonary aspergillosis. Intensive Care Med. 2011;37:1605e12.

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