Al-Qadisiyah Journal of Pure Science

Manuscript 1274

The Co-infection of pulmonary Mycosis with Tuberculosis among Iraqi patients: A cross-sectional study

Mahmood Yaseen Mukhlif

Yassir M. Abdulateef

Sawsan Qahtan Taha Al-Quhli

ORIGINAL STUDY

The Co-infection of Pulmonary Mycosis With Tuberculosis Among Iraqi Patients: A Cross-sectional Study

Mahmood Y. Mukhlif*, Yassir M. Abdulateef, Sawsan Q.T. Al-Quhli

Department of Microbiology, College of Medicine, University of Anbar, Anbar, Iraq

Abstract

Background and aim of study: **Pulmonary tuberculosis is a serious airborne-transmitted infectious disease that is caused by significant threat pathogenic bacteria known as *Mycobacterium tuberculosis* and is amongst the top ten of the deadliest single infectious agents globally. Pulmonary mycosis is a lung fungal infection that is common among patients who suffer from immune system suppression. PTB and PM have exhibited similar or coordinated risk factors. Furthermore, recent studies have assumed PTB as a significant PM risk factors. Pulmonary tuberculosis patients were frequently suggested to have pulmonary fungal coinfection that increased the rate of mortality. This study aimed to assess the pulmonary mycosis co-infection prevalence in pulmonary tuberculosis Iraqi patients.**

Materials and methods: **A cross-sectional study carried out from February to July 2023. The study included 150 participants from patients suspected of having pulmonary tuberculosis. Two early morning sputum samples were obtained from each participants. Tuberculosis has been identified using GeneXpert system, whereas mycotic infections have been detected using conventional methods including KOH, culture, and Lactophenol blue stain.**

Results: **Among 150 Iraqi participants 66.7% (100/150) were positive for PTB, 60% (90/150) were positive for fungal pathogen. Pulmonary tuberculosis fungal co-infection among PTB patients was seen in 81% (81/100). *Candida* was the predominant species participating in pulmonary mycosis. There was a statistically significant association between pulmonary mycosis and PTB ($p < 0.0001$).**

Conclusion: **The study found that PTB Iraq patients exhibited a higher pulmonary fungus coinfection prevalence, *Candida* is the main pathogenic fungus in pulmonary mycosis, *C. albicans* being the predominant species.**

Keywords: Tuberculosis, *Mycobacterium tuberculosis*, Pulmonary mycosis, Fungal co-infections, *Candida*, *Aspergillus*

1. Introduction

Pulmonary tuberculosis (PTB) is a potentially fatal contagious disease caused by significant threat pathogenic bacteria known as *Mycobacterium tuberculosis* (*Mtb*) which mostly attacks the lungs. PTB ranks among the top 10 global causes of mortality. The World Health Organization (WHO) estimated ten million individual developed PTB, resulting in 1.5 m deaths annually [1]. 95% of cases of tuberculosis were reported in developing countries, especially in Asia, Africa, the Middle, where diagnostic and treatment facilities are limited [2]. PTB is an airborne disease, transmitted by inhalation of droplets from infected individuals by cough, talk, singing, and sneezing [3]. PTB symptoms are unspecific, and include chest pain, cough with bloody sputum, fatigue, fever, hemoptysis, and dyspnea [4]. As a result of immunocompromised, changes in the structure of bronchial, and damage in lung tissue, patients with PTB are susceptible to combination infections, particularly pulmonary fungal infections [5].

Pulmonary mycosis (PM) is a lung fungal infection that occurs when fungal spores are inhaled, resulting

Received 16 November 2023; accepted 11 April 2024.
Available online 20 August 2025

* Corresponding author.
E-mail addresses: mahmood.vb43@gmail.com (M.Y. Mukhlif), dr.yasserjanai@uoanbar.edu.iq (Y.M. Abdulateef), sawsanqt@uoanbar.edu.iq (S.Q.T. Al-Quhli).

https://doi.org/10.29350/2411-3514.1274

in the invasion of the bronchial and lung tissues and the development of inflammatory lesions within the lungs. Patients with immunodeficiency diseases like HIV/AIDS and those receiving immunosuppressive therapy, such as those undergoing bone marrow/stem cell transplantation, are most likely to develop it [6,7]. According to WHO, Out of a total of 13 million fungal infections reported cases worldwide, resulting in 1.5 million deaths, approximately 60% were attributed to PM [8]. PM may be caused by either endemic fungi, opportunistic fungi, or a combination of both opportunistic and endemic fungi. Endemic fungus like *Candida albicans*, *Aspergillus fumigatus*, and *Trichophyton rubrum*. Opportunistic fungus like *Aspergillus species, Cryptococcus species, Pneumocystis species, and Histoplasma species* [9].

Fungal co-infections play a vital role in individuals with immunodeficiency disorders like severe pulmonary diseases or AIDS, who are being treated with antibiotics and/or corticosteroids. Numerous studies conducted globally have documented the occurrence of pulmonary fungal coinfections in patients with PTB, resulting in a significant mortality rate. Up to one million people who recover from PTB develop lung fungal infections every year [10]. PTB and PM have exhibited similar or coordinated risk factors. Old age, immunosuppressed states, administration of corticosteroids or other immunosuppressive medications have been reported as a common risk factors for PTB and the same with PM [8,11]. Furthermore, recent studies have assumed PTB as a significant PM risk factors [12,13]. The severity of fungal co-infections and prolonged duration of treatment require sufficient attention to this issue. So, the aim of this study is to assess the pulmonary mycosis co-infection prevalence in pulmonary tuberculosis Iraqi patients.

2. Materials and methods

This cross-control study included 150 participants from patients suspected of having pulmonary tuberculosis aged from 17 to 73. The participants have matched age and sex. Three hundred sputum samples were collected from patients: two samples from everyone, one for tuberculosis identification, and the other for fungal infection identification. Patients were selected from those admitted to the University of Anbar in Al-Anbar Governorate, Iraq between Mar 2023 and July 2023 under ethical committee of the hospital. The study was approved by the university research ethics committee (No. 195, Date: 20-2-2023). Written informed consent of all patients was obtained before sputum sample collection.

2.1. Identification of tuberculosis

Morning two sputum specimens from each patient were collected in a in specific container and stored at 4–8 °C till diagnoses. *Mtb* was detected using the GeneXpert MTB/RIF assay (Cepheid, Sunnyvale, USA). The Sputum sample and reagent were mixed at room temperature. The mix was incubated for 2 h into the GeneXpert system. The outcome was subsequently interpreted either "MTB detected" or "MTB not detected." PTB diagnosis was confirmed with conventional methods including chest X-ray and clinical diagnosis of symptoms.

2.2. Identification of fungi

Sputum samples were cultured on Sabouraud dextrose agar (SDA) (HiMedia; India) containing supplemented with 20 μg/ml of chloramphenicol (SC). The plates incubated at 25 °C. After seven days, the fungal growth was inspected, before being reported, the growing fungal colonies were kept under observation for four weeks. Samples were reported as negative only if there was no growth after four weeks of incubation. The positive colonies were classified by their macroscopic and microscopic characteristics after Lactophenol blue stain. The smooth, white pasty colonies refer to *Candida* spp. and rough, greenish, brown-pigmented colonies refer to *Aspergillus* spp.

2.3. Inclusion and exclusion criteria

Patients with clinical pulmonary tract infection, particularly of those with a persistent cough for more than three weeks were included in the study. Individuals who were under antifungal treatment or failed to provide samples were excluded.

2.4. Statistical analysis

Data Analysis was performed using SPSS program version 27. Categorical variables are expressed as frequencies and percentages. Chi square test ($\chi 2$) was used for comparison between categorical variables. P value < 0.05 was statistically significant.

Table 1. Demographic data of participants.

		Positive Tuberculosis N = 100		Negative Tuberculosis N = 50		P value
		No	%	No	%	
Age (years)	<20 years	9	9	4	8	0.096 **ns**
	20–29	15	15	14	28	
	30–39	30	30	20	40	
	40–49	16	16	7	14	
	50–59	13	13	2	4	
	60–69	9	9	2	4	
	≥70 years	8	8	1	2	
	Mean ± SD (Range)	40.7 ± 13.1 (19–73)		36.1 ± 9.8 (17–71)		
Gender	Male	70	70	30	60	0.297 **ns**
	Female	30	30	20	40	

P value of Pearson Chi-square test at 0.05 level.

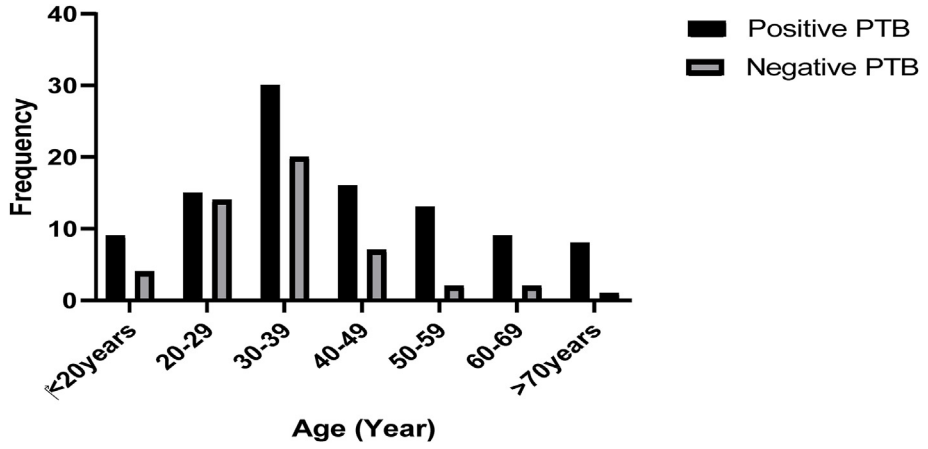

Fig. 1. Age of the study subjects.

3. Results

This study includes one hundred fifty (150) participants of which 100 (66.7%) were males and 50 (33.3%) were females. The study subjects' age varied from 17 to 73 years with 40 years as mean age. Age and sex were matched between participants. PTB was diagnosed in 66.7% (100/150) sputum samples of patients while 33.3% (50/150) samples showed negative PTB (Table 1) and Fig. 1.

Among confirmed PTB patients, 19 (19%) of fungal culture showed no growth, fungal species were isolated from 81 samples, among them 49% showed *candida* species which were the most prevalent species; the *C. albicans* (33%), *C. tropicalis* (6%), *C. krusei* (5%), *C. glabrata* (4%), *C. parapsilosis* (1%), 35% showed *Aspergillus* species; *Asp. Fumigatus* (17%), *Asp. Niger* (8%), *Asp. Flavus* (7%), *Asp. Nudilans* (3%), 4% showed Cryptococcus *neoformans* us species, and 1% showed *Alternaria alternata* species. Among 50 Non-PTB individuals, 9 (18%) samples were positive for *C. albicans*, and 41 (82%) samples showed no growth as shown in Table 2 and Fig. 2.

There is a statistically significant association between PTB disease and pulmonary mycosis coinfections prevalence in the study subjects Table 3 and Fig. 3.

Table 2. Fungai isolates from PTB patients.

Fungal	No	%	Total
No growth	19	19	19%
Candida albicans	33	33	49%
Candida tropicalis	6	6	
Candida krusei	5	5	
Candida galbrata	4	4	
Candida parapsilosis	1	1	
Aspergillus fumigatus	17	17	35%
Aspergillus niger	8	8	
Aspergillus flavus	7	7	
Aspergillus nudilans	3	3	
Cryptococcus neoformans	4	3.7	4%
Alternaria alternata	1	1	1%

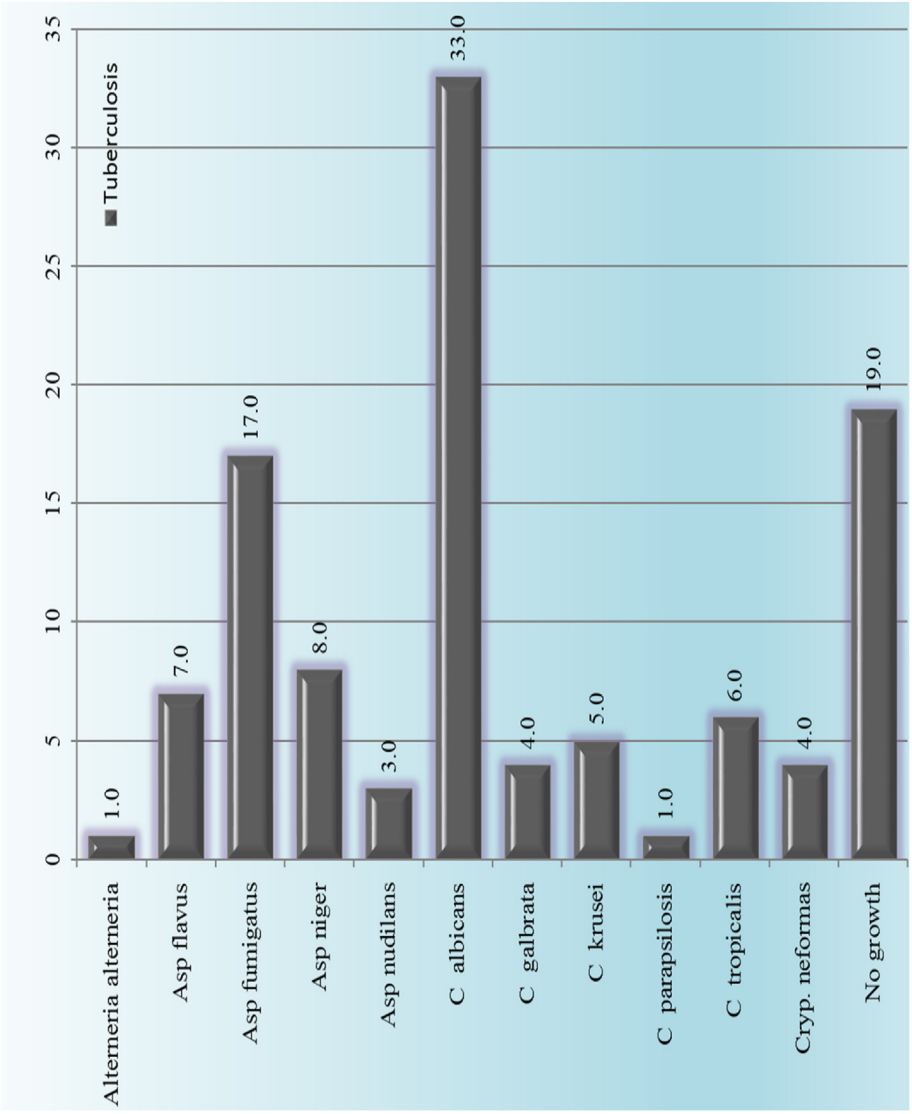

Fig. 2. Types of candida isolated from PTB patients.

4. Discussion

Pulmonary tuberculosis (PTB) is among the most harmful infectious disease that caused by *Mycobacterium tuberculosis* (*Mtb*), *Mtb* is a fatal bacteria that spread from infected one to another by air droplet through cough and sneezing and causes thousands deaths worldwide especially in developing countries [14]. Pulmonary mycosis is a respiratory fungal infection disease resulting from various pathogens like *C. albicans* or *Asp. fumigatus* [15]. Several global reports have revealed the occurrence of pulmonary fungal coinfections in patients with PTB that increased the rate of morbidity and mortality due to

Table 3. Prevalence of tuberculosis, fungal mycosis, and coinfections among study subjects.

		Fungal		X^2	P. value
		Positive	Negative		
PTB	Positive	81	19	52.531	<0.0001
	Negative	9	41		

P value of Pearson Chi-square test at 0.05 level.

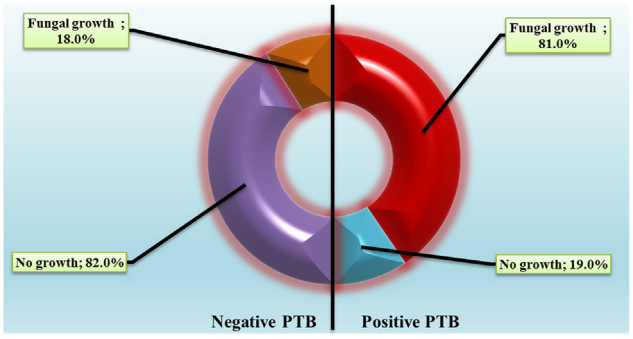

Fig. 3. Fungal growth among the studied subjects.

the lack of diagnosis and management especially in developing regions, where there is low awareness of pulmonary mycosis risk [16]. The aim of this study is to assess the pulmonary mycosis co-infection prevalence in pulmonary tuberculosis Iraqi patients.

The current study found a higher prevalence rate of PTB among Iraq population that found to be 66.6%. This finding was similar to that reported by Al-Hadraawy et al., [17] who record the percentages of patients infected with PTB in AL-Najaf Governorate, Iraq. The study reported that PTB prevalence in the north section of the governorate was 58% and in the south section was 42%, the results demonstrated higher PTB prevalence among Iraqi population which could be a result of the growing population and the numerous populated and rural places. Also, Our finding agreed with another study in Iraq by Aljanaby et al., [18], showed that high prevalence rate of PTM infection in Baghdad; in AL-Karkh side was 35.4% and in AL-Rusafa side was 64.6%.

Tuberculosis carries on being a serious public health concern especially in low and middle-income country. WHO reports that nine million new PTB cases are recorded annually, Asia and Africa representing the majority of them? WHO documented a high PTB prevalence rate in the Eastern Mediterranean countries, among these countries Iraq where tuberculosis is more common [19], this may be due to delay in diagnosis and treatment administration. Early diagnosis and accurate identification of TB infection is the key for prevention of the disease. Low rates of detection may be due to conventional approaches to diagnosis that have major drawbacks. Rapid, accurate, inexpensive test for PTB diagnosis has become a critical to provide an effective treatment [20].

In this study prevalence of pulmonary fungal infection and PTB-pulmonary mycosis coinfection was found to be 60%, and 81%, respectively. Our study showed a higher prevalence of pulmonary fungal coinfection. Similar to our findings, Mortazavi et al., [21] observed a significant incidence of fungus-TB co-infection in Iran. The study found that according to TB patients, the prevalence of fungus-TB co-infection ranged from 12.3 to 68.8%. Our findings also are consistent with published results from other population based studies by Talle et al., [22] who reported a higher fungal infection incidence 68% in patients with PTB, as well as higher prevalence fungal coinfections 90.2% among participants coinfected TB in Nigeria. In addition, Danlami et al., [8] found a significant pulmonary fungal incidence 71.2%, PTB patients had a moderate incidence of PM coinfection 29.4%. The area, population, design of the study as well as the method of diagnosis may be the reasons responsible for the prevalence difference.

Numerous reasons are thought to be contributing to the global rise in fungal infections, but PTB and widespread use of immunosuppressive medications, antibiotics, and steroids continue to be the main cause. PTB patients are mostly immunocompromised, and more susceptible to fungal infection, thus decreasing host immunity, increase the virulence of the tuberculosis infection, making it challenging to treat [23].

The current investigation discovered that *Candida* spp. was the most prevalent fungus associated with co-infection with TB, with a prevalence rate of 49%, followed by *Aspergillus* spp. 35%. Additionally, *C. albicans* was the most prevalent fungus 33% among the *Candida* spp., occurring 33% of the time. The current study findings are consistent with the investigation of the reports of Danlami et al., [8] in Nigeria, Hussein et al., [16] in Iraq, and Mortazavi et al., [21] in Iran whose studies showed that *Candida* spp. was the most prevalent fungus associated with co-infection with TB followed by *Aspergillus* spp. In disagreement with our findings, reports from different population by Hosseini et al., [24], Amiri et al., [2], and Aghili et al., [25] who found *Aspergillus* spp. was the most frequent and Asp. fumigatus was the predominant spp. in pulmonary coinfected patients.

In this current study, there is a statistically significant association between prevalence of PTB disease and pulmonary mycosis coinfections among study subjects. Compared to non-PTB patients, PTB patients exhibited a greater prevalence of lung fungal infection, our results suggest that PTB patients have higher susceptibility for pulmonary fungal infection. Our finding is agreed with study by Talle et al., [22] who found a statistical relation between relationship and TB ($P < 0.05$), indicated that lung fungal infections were more common and prevalent in TB patients 80% compared with those without TB 10%. Also, in line with our findings Amiri et al., [2] indicated that there is a statistically significant correlation between PTB and the occurrence of fungi coinfection (P value < 0.05) and suggested that tuberculosis could be a risk factor for fungal infection. As well as, Hadadi-Fishani et al., [10], who reported that PTB patients had a significant rate of fungus coinfection., and hypothesized that patients with PTB have impaired immune systems, making them more vulnerable to fungal and pulmonary mycotic infections.

There is a relatively high incidence of fungal infection among tuberculosis cases The occurrence of fungal coinfection concomitantly with tuberculosis is

of paramount interest in the treatment and management of PTB patients as *C. albicans* is supposed to enhance the virulence of *Mtb*, this coinfection increase the enormous global burden of PTB and the overall rates of morbidity and mortality [26].

5. Conclusion

In conclusion, PTB Iraq patients exhibited a higher pulmonary fungus coinfection prevalence, *Candida* is the main pathogenic fungus in pulmonary mycosis, *C. albicans* being the predominant species.

Funding

Self-funding.

References

[1] Asra'a A, Al-Khafaji ZM, Al-Ouqaili MT. Investigation of Accd3 gene of *Mycobacterium tuberculosis* Iraqi isolates. Asian J Pharmaceut Clin Res 2018;11(8):208–11.
[2] Amiri MRJ, Siami R, Khaledi A. Tuberculosis status and coinfection of pulmonary fungal infections in patients referred to reference laboratory of health centers ghaemshahr city during 2007–2017. Ethiop J Health Sci 2018 Nov; 28(6):683–90.
[3] Eyad HN, Abdulateef YM. Abnormal presentation of tb patients: anthropological study. Ann Trop Med Publ Health 2019;22:46–55.
[4] Aljanabi YM, Lafi SA, Eyada HN. Tb laboratory diagnosis, a comparative study in Baghdad, Iraq. Prof(Dr) RK Sharma 2020;20(4):4885.
[5] Yan H, Guo L, Pang Y, Liu F, Liu T, Gao M. Clinical characteristics and predictive model of pulmonary tuberculosis patients with pulmonary fungal coinfection. BMC Pulm Med 2023;23(1):56.
[6] Ekeng BE, Davies AA, Osaigbovo II, Warris A, Oladele RO, Denning DW. Pulmonary and extrapulmonary manifestations of fungal infections misdiagnosed as tuberculosis: the need for prompt diagnosis and management. J Fungi 2022 Apr 28;8(5):460.
[7] Liu A, Li Z, Su G, Li Y, Zhang Y, Liang J, et al. Mycotic infection as a risk factor for COVID-19: a meta-analysis. Front Public Health 2022 Sep 7:10.
[8] Danlami MB, Adefowepo AM, Manga SS, Yahaya TO, Mshelia MB, Kalgo ZM. Pulmonary mycoses among pulmonary tuberculosis in Kebbi state North Western Nigeria. Egypt J Bronchol 2023;17(1):1–7.
[9] Lin J, Feng B, Tang H, Xu H, Tang Y. A systematic review and meta-analysis: pulmonary mycosis pathogen distribution. Ann Palliat Med 2021;10(7):7919–32.
[10] Hadadi-Fishani M, Shakerimoghaddam A, Khaledi A. Candida coinfection among patients with pulmonary tuberculosis in Asia and Africa; A systematic review and meta-analysis of cross-sectional studies. Microb Pathog 2020 Feb; 139:103898.
[11] Sani FM, Uba A, Tahir F, Abdullahi IN, Adekola HA, Mustapha J, et al. Spectrum of pulmonary fungal pathogens, associated risk factors, and anti-fungal susceptibility pattern among persons with presumptive tuberculosis at Gombe, Nigeria. Int J Mycobacteriol 2020;9(2):144–9.
[12] Sharma A, Goel A. Mucormycosis: risk factors, diagnosis, treatments, and challenges during COVID-19 pandemic. Folia Microbiol (Praha) 2022 Jun 26;67(3):363–87.
[13] Prakash H, Ghosh AK, Rudramurthy SM, Singh P, Xess I, Savio J, et al. A prospective multicenter study on mucormycosis in India: epidemiology, diagnosis, and treatment. Med Mycol 2019;57(4):395–402.
[14] Gopalaswamy R, Shanmugam S, Mondal R, Subbian S. Of tuberculosis and non-tuberculous mycobacterial infections–a comparative analysis of epidemiology, diagnosis and treatment. J Biomed Sci 2020;27(1):1–17.
[15] Muhammad Sani F, Abdullahi IN, Sunday Animasaun O, Elisha Ghamba P, Umar Anka A, Oluwafemi Salami M, et al. Prevalence and risk factors of pulmonary fungal pathogens among symptomatic patients with or without tuberculosis at Gombe, Nigeria. J Med Microbiol Infect Dis 2020;8(3):76–83.
[16] Hussein HM, Sekhi AA, Sekeb HS. Prevalence of fungi in clinically suspected cases of pulmonary tuberculosis in Iraq, Wasit. Sys Rev Pharm 2021;12(1):1393–6.
[17] Al-Hadraawy SK, Alhadrawi KK, Aljanaby IAJ, Aljanaby AAJ, Zabibah RS. Prevalence of pulmonary tuberculosis in Al-Najaf governorate, Iraq. F1000Research 2022;11: 675.
[18] Aljanaby AAJ, Al-Faham QMH, Aljanaby IAJ, Hasan TH. Epidemiological study of *Mycobacterium tuberculosis* in Baghdad governorate, Iraq. Gene Rep 2022;26:101467.
[19] Karadakhy K, Othman N, Ibrahimm F, Saeed AA, Amin AA-AH. Tuberculosis in sulaimaniyah, iraqi kurdistan: a detailed analysis of cases registered in treatment centers. Tanaffos 2016;15(4):197.
[20] Al-Ouqaili MTS. Molecular detection of insertion sequence 6110 of *Mycobacterium tuberculosis* in patients with pulmonary tuberculosis and tuberculous pleuritis in Anbar Governorate, West of Iraq. Int J Life Sci Pharma Res 2018;8(3): 46–57.
[21] Mortazavi H, Ghazalibina M, Mansouri S, Khaledi A, Saburi E. Pulmonary fungal co-infection prevalence among Iranian patients with pulmonary tuberculosis: a systematic review and meta-analysis. Sains Malays 2019;48(12):2717–25.
[22] Talle M, Hamidu IM, Nasir I-A, Mursal A, Dikwa KB, Jelili M, et al. Prevalence and profile of pulmonary fungal pathogens among HIV-infected patients attending University of Maiduguri Teaching Hospital, Nigeria. Egypt J Intern Med 2017;29(1):11–5.
[23] Muni S, Rajpal K, Kumar R, Kumari R, Sinha R, Kumar S, et al. Identification of fungal isolates in patients with pulmonary tuberculosis treated at a Tertiary care Hospital. Cureus 2023;15(4):e37664.
[24] Hosseini M, Shakerimoghaddam A, Ghazalibina M, Khaledi A. Aspergillus coinfection among patients with pulmonary tuberculosis in Asia and Africa countries; a systematic review and meta-analysis of cross-sectional studies. Microb Pathog 2020;141:104018.
[25] Aghili SR, Shokohi T, Hedayati MT, Abastabar M, Aliyali M, Hasanpour H. Invasive forms of Candida and Aspergillus in sputum samples of pulmonary tuberculosis patients attending the tuberculosis reference laboratory in Ghaemshahr, Northern Iran: an analysis of samples collected during the past 10 years. Int J Mycobacteriol 2016;5(1):S179–80.
[26] Mitalkumari G, Prajapati K, Vegad MM. Candida-Co infection: prevalence in pulmonary tuberculosis patients a tertiary Care Hospital, Ahmedabad. Int J Contemp Microbiol 2019; 5(2):1–5.