http://ijmpes.com
doi:10.34172/ijmpes.6301
Open Access
Original Article
APMI Press
Aras Part Medical International Press
A scientific publishing imprint of PMC Pharmaceutique France
Int J Med Parasitol Epidemiol Sci 2026;7(Suppl 1):S63-S69
eISSN 2766-6492
International Journal of Medical Parasitology & Epidemiology Sciences

Serum Tumour Necrosis Factor-Alpha and Interleukin-4 as Immunological Indicators of hwp1-Confirmed *Candida albicans* Respiratory Tract Infection: A Case-Control Study from Wasit Province, Iraq

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Abstract

Introduction: *Candida albicans* is a critical-priority fungal pathogen and an increasingly recognised cause of respiratory tract infection in hospitalised and immunocompromised patients. Tumour necrosis factor-alpha (TNF-α) and interleukin-4 (IL-4) exert opposing influences on antifungal immunity, yet their combined behaviour in molecularly confirmed respiratory candidiasis remains poorly defined. Aim of this study is to quantify serum TNF-α and IL-4 in Iraqi patients with hwp1-confirmed *C. albicans* respiratory infection and to evaluate their diagnostic performance.

Methods: A case-control study enrolled 200 patients with respiratory tract infection and 100 age- and sex-matched healthy controls at three facilities in Al-Kut, Wasit province, Iraq, between December 2025 and April 2026. Isolates were identified by conventional mycology and the VITEK®2 Compact system, then confirmed by polymerase chain reaction targeting the hwp1 gene. Serum cytokines were measured by sandwich ELISA, and diagnostic accuracy was assessed by receiver operating characteristic analysis.

Results: *C. albicans* was cultured from 42 patients (21.0%) and from no control ($P<0.0001$); 37 isolates (88.1%) yielded the 941 bp hwp1 amplicon. TNF-α was markedly higher in patients (156.6 ± 38.20 versus 34.85 ± 14.73 ng/L) as was IL-4 (56.59 ± 17.19 versus 17.78 ± 6.783 ng/L; both $P<0.0001$). Both cytokines achieved an area under the curve of 1.000 with 100% sensitivity and specificity. TNF-α correlated inversely with procalcitonin among patients ($r=-0.339$, $P=0.028$).

Conclusion: Respiratory candidiasis is accompanied by simultaneous pro-inflammatory and Th2 activation. Combined measurement of TNF-α and IL-4 with hwp1 detection warrants evaluation as a diagnostic adjunct.

Keywords: *Candida albicans*, Respiratory tract infection, Tumour necrosis factor-alpha, Interleukin-4, hwp1 gene, Iraq

Received: April 19, 2026, **Revised:** June 3, 2026, **Accepted:** July 28, 2026, **ePublished:** August 23, 2026

Introduction

Fungal disease has emerged as a substantial and historically underestimated contributor to global mortality. Contemporary burden estimates indicate approximately 6.5 million invasive fungal infections and 3.8 million associated deaths annually, of which roughly 2.5 million are directly attributable to the fungal process itself; *Candida* bloodstream infection and invasive candidiasis alone account for an estimated 1,565,000 cases and 995,000 deaths each year (1). In recognition of this burden and of escalating antifungal resistance, the World Health Organization designated *Candida albicans* a critical-priority fungal pathogen in its inaugural fungal priority pathogens list, placing it alongside *Aspergillus fumigatus, Candidozyma auris* and *Cryptococcus neoformans*(2,3). The regional dimension of this problem is equally pronounced. Reported candidemia incidence reaches 15.4 per 100,000 population in Qatar and 11.8 per 100,000 in the United Arab Emirates, with 49.2% of the latter occurring in intensive care units, while nosocomial candidiasis in Türkiye ranges from 1.2 to 5.6 per 1,000 admissions and invasive candidiasis in Saudi Arabia reaches 26 cases per 1,000 intensive care admissions(4). Comparable surveillance data from Iraq remain scarce, and information specific to the respiratory tract is scarcer still.

Among the fungal pathogens capable of causing respiratory disease, *C. albicans* holds a prominent position, particularly in patients whose immune defences

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have been compromised by underlying illness, prolonged hospitalisation, or immunosuppressive therapy (5,6). Its clinical relevance in this context is reinforced by recent epidemiological analysis, in which pulmonary disease was the most frequent underlying comorbidity among adults with candidiasis, present in 47.8% of cases, and in which respiratory dysfunction emerged as an independent predictor of thirty-day mortality(7). Nevertheless, *Candida* frequently colonises the respiratory tract without causing harm, and its transition to a genuine pathogen in susceptible hosts can produce serious infection with appreciable morbidity(8). Distinguishing this transition from innocent colonisation remains one of the central challenges in managing respiratory infection in high-risk clinical settings, and it is precisely this diagnostic ambiguity that motivates the search for host-derived markers capable of signalling true invasive disease.

Beyond the difficulty of separating infection from colonisation, the host immune response to this fungus is complex and incompletely characterised. At the heart of antifungal host defence lies a cytokine network that coordinates recognition, effector recruitment and resolution. Tumour necrosis factor-alpha (TNF-α) occupies a central and largely protective position within this network. Produced by macrophages and monocytes upon recognition of fungal cell wall β-glucan and mannan through Dectin-1 and Dectin-2 receptors, TNF-α promotes phagocyte activation, supports neutrophil recruitment to infected tissue, and sustains the Th1-polarised responses required for durable antifungal protection(9). The functional necessity of this axis is demonstrated clinically by the substantially elevated risk of invasive candidiasis in patients receiving anti-TNF biologics or harbouring congenital defects in TNF-α signalling (10).

Interleukin-4 (IL-4) operates as a functional counterweight to TNF-α and to the broader Th1 axis. As the canonical Th2-inducing cytokine, IL-4 suppresses macrophage activation, inhibits pro-inflammatory cytokine production, and directs the immune response toward humoral rather than cell-mediated mechanisms(11). In the setting of fungal infection this Th2 bias is largely counterproductive, since IL-4 reduces macrophage killing capacity against *C. albicans* and impairs oxidative burst responses, thereby creating conditions that favour fungal persistence over clearance(12). Its elevation in infected patients may therefore reflect not a beneficial adaptation but an immunological misdirection that coexists with, and partially undermines, the TNF-α-driven pro-inflammatory effort. Examining these two mediators together consequently offers a more informative picture of the immunological state of respiratory candidiasis than either would provide in isolation.

Confirming the species identity of respiratory *Candida* isolates at the molecular level adds essential specificity to such investigation. Polymerase chain reaction targeting the hwp1 gene, which encodes hyphal wall protein 1, a mannoprotein expressed exclusively during hyphal morphogenesis of *C. albicans* and absent from closely related species such as *C. dubliniensis*, provides a rapid and reliable method for species-level confirmation (13,14). The characteristic 941 bp amplicon corresponds to the Class IV homozygous genotype of *C. albicans*(13). Despite the recognised roles of TNF-α and IL-4 in fungal immunopathology, published data on their serum concentrations in patients with molecularly confirmed respiratory *C. albicans* disease remain limited, and no study has addressed this question in the population of Wasit province, Iraq. The present work was undertaken to address this gap by measuring serum concentrations of both cytokines in confirmed patients and healthy controls, evaluating their diagnostic accuracy, and examining their correlations with other measured parameters within the patient group.

Patients and Methods

Study Design, Setting, and Ethics

Between December 2025 and April 2026, a case-control study was conducted at three facilities in Al-Kut city, Wasit province, Iraq: Al-Zahraa Teaching Hospital, Al-Karama Teaching Hospital, and Respiratory Diseases Clinic. The study was approved by the Institutional Research Ethics Committee (IREC) of the College of Medicine, University of Thi-Qar (Approval ID: IQ.UTQ.MED.2024.030, date: 17\11\2025). Before any study procedure was initiated, all participants or their legal guardians where applicable received a clear explanation of the study's purpose and provided informed consent. All procedures were carried out in compliance with the principles of the declaration of Helsinki.

Participants

Two hundred consecutive patients presenting with respiratory tract infection symptoms including productive cough, fever, dyspnea, or abnormal chest findings were assessed for enrollment. Inclusion required that the participant provide an adequate, uncontaminated respiratory specimen. Exclusion criteria comprised: ongoing antifungal therapy at the time of specimen collection; concurrent pulmonary tuberculosis; known autoimmune disease or hematological malignancy; inadequate specimen quality (salivary sputum with > 25 squamous epithelial cells per low-power field); and pregnancy. One hundred healthy volunteers matched by age group and sex were enrolled as controls from individuals attending routine outpatient check-up visits at the same institutions. Controls had no active illness, no history of recent infection, and were not receiving any medications at the time of enrollment.

Specimen Collection and Mycological Identification

Respiratory specimens were collected according to clinical presentation: throat swab in Amies transport medium for patients with upper respiratory involvement; deep-cough early morning sputum in sterile wide-mouth containers for those with lower respiratory symptoms; and Bronchoalveolar lavage (BAL) fluid obtained by flexible bronchoscopy, performed by qualified pulmonologists,

from ICU patients. All specimens were transferred to laboratory and processed within two hours of collection. Specimens were inoculated onto Sabouraud Dextrose Agar (SDA) supplemented with chloramphenicol and incubated at 37 ºC for 24 to 48 hours. Yeast colonies were sub-cultured on HiCrome Candida Differential Agar (Himedia, India) for presumptive species differentiation based on colony color. Isolates producing characteristic light-green colonie characteristic of *C. albicans* on this medium. Further confirmation was achieved by Gram staining, Lactophenol Cotton Blue (LPCB) staining, and Germ Tube test in human serum at 37 ºC for 2-3 hours. Final species confirmation was performed using the Vitek® 2 Compact System (bioMèrieux, France).

Molecular Confirmation

DNA was extracted from all 42 culture-confirmed *Candida albicans* isolates using the Magen HiPure Yeast DNA kit (Magen Biotechnology Co., Ltd., Guangzhou, China), following the manufacturer's protocol. The kit employs silica column purification without phenol or chloroform, with total extraction completed within 60 minutes. Molecular confirmation was carried out by conventional PCR targeting the hyphal wall protein1(hwp1) gene, a mannoprotein expressed exclusively during hyphal morphogenesis of *C. albicans* and absent in closely related species(13,14). The species-specific primers, as described by Klesiewicz et al.(13), were used as presented in (Table 1)

Polymerase chain reaction (PCR) was performed in final reaction volume of 25 µL. Each reaction contained 12.5 µL of 2 × PCR Master Mix (ELK Biotechnology, China), 5 µL of template DNA, 2 µL each of forward and reverse primers (Macrogen Inc., Seoul, South Korea), and 3.5 µL of nuclease-free water (Promega, USA). Amplification was performed under optimized thermal cycling conditions specific for target gene, as outlined in (Table 2).

Amplification products were resolved by electrophoresis on a 1.5% agarose gel stained with ethidium bromide and visualized under ultraviolet illumination. A single band at 941 base pair (bp) was interpreted as a positive result for *C. albicans,* consistent with the Class IV homozygous HWP1 genotype(13). hwp1 PCR was applied exclusively to culture-positive isolates; healthy controls, being culture-negative, were not subjected to molecular analysis.

Table 1. The name, sequence, and product size of primers used in this study

Gene	Primer Sequence	Product Size (bp)
hwp1	F:5'- GCTAATGGAAGCTTTGATTCTAATG-3' R:5'-AAGTATGGAGTTGTGGCAATGAAATC-3'	941 bp

Table 2. PCR Thermal cycling conditions for (hwp1) gene amplification

CycleNo.	Stage	Temperature (ºC)	Time
1	Initial denaturation	94	4 min
	Denaturation	94	10 s
10 ×	Annealing	54	30 s
	Extension	68	45 s
	Denaturation	94	15 s
25 ×	Annealing	54	30 s
	Extension	68	45 s
1	Final extension	68	7 min
1	Cooling	4	10 min

ELISA Measurement of Serum TNF-α and IL-4

Five milliliters of venous blood were drawn aseptically from each participant using sterile vacutainer tubes. Samples were allowed to clot at room temperature for 30 minutes, then centrifuged at 3000 rpm for 10 minutes. Separated serum was aliquot and stored at −20 ºC until the day of analysis. The serum concentrations of TNF-α and IL-4 (expressed in ng/L) were determined using commercially available sandwich ELISA kits (BT LAB, China) employed the End point method and non-linear regression calculation (correlation coefficients: r = 0.978 for TNF-α and r = 0.986 for IL-4; detection ranges: 0–960 ng/L for TNF-α and 0–1280 ng/L for IL-4). All assay steps were performed according to manufacturer's protocol. Optical densities were read at 450 nm using a microplate reader, and cytokine concentrations were derived from standard calibration curves.

Statistical Analysis

Data were analyzed using GraphPad Prism version 10.5.1 (GraphPad Software, LLC, San Diego, CA, USA). Categorical variables were expressed as frequency and percentage (No., %) and compared between groups by Chi-square (χ^2) test. Continuous variable were expressed as mean ± standard deviation (Mean ± SD) and compared by independent samples t-test. Receiver operating characteristic (ROC) curve analysis was performed to assess the diagnostic accuracy of serum TNF-α and IL-4, reporting the area under the curve (AUC), optimal cut-off value, sensitivity, and specificity. Correlation between continuous parameters within the patient group were assessed using Pearson's correlation coefficients. A P -value of ≤ 0.05 was considered statistically significant for all analyses.

Results

Demographic Characteristics

The two groups were adequately matched for both age and sex. The mean patient age was 49.26 ± 18.25 years, compared with 53.36 ± 16.96 years in controls ($P = 0.061$, NS). Female participants were slightly more frequent in both groups: 106 (53.0 %) among patients and 59 (59.0 %) among controls, with no significant difference in sex distribution ($P = 0.74$). These findings confirm that any observed differences in cytokine levels are attributable to infection status rather than demographic confounding. Full demographic data are presented in Table 3.

Candida albicans Isolation and hwp1 Molecular Detection

Candida albicans was isolated by culture from 42 of the 200 enrolled patients (21.0%), while none of the healthy controls yielded a positive culture result ($\chi^2 = 22.70$; $P < 0.0001$). Molecular analysis of the 42 culture-confirmed

isolates using hwp1 PCR produced expected 941 bp amplicon — indicative of the Class IV homozygous HWP1 genotype — in 37 cases (88.1%; Lanes 1_5, 7_14, 16_27, 29_ 36, 38_41). The remaining five isolates (11.9%; Lanes 6, 15,28,37 and 42) yielded no amplification product despite culture-positive most likely reflecting phenotypic misidentified non-albicans *Candida* species at culture stage, a limitation previously reported with chromogenic agar-based identification(8). hwp1 PCR was applied exclusively on culture-positive isolates; healthy controls, being culture-negative, were not subjected to molecular analysis. Gel electrophoresis results are shown in Figure 1 and summarized in Table 4.

Serum TNF-α and IL-4 Levels.

Serum concentrations of both cytokines were substantially elevated in confirmed *Candida* patients (n = 42) than in healthy controls (n = 100). Mean TNF-α in patients reached 156.6 ± 38.20 ng/L against 34.85 ± 14.73 ng/L in controls — a roughly 4.5 fold difference ($P < 0.0001$). Mean IL-4 was 56.59 ± 17.19 ng/L in patients and 17.78 ± 6.783 ng/L in controls, reflecting a more than three-fold elevation ($P < 0.0001$). Within the confirmed patient group, a statistically significant negative correlation was found between TNF-α and procalcitonin (r = −0.339, P = 0.028); no other inter-parameter correlations reached significance. These data presented in Table 5.

Diagnostic Accuracy by ROC Curve Analysis

ROC analysis demonstrated that both cytokines (TNF-α and IL-4) achieved an AUC of 1.000 ($P < 0.0001$), indicating perfect discrimination between confirmed *Candida* patients and healthy controls. TNF-α at a cut-off of > 71.21 ng/L and IL-4 at a cut-off of > 29.75 ng/L each achieved 100.0% sensitivity and 100.0% specificity (Table 6).

Discussion

The present study demonstrates that serum concentrations of TNF-α and IL-4 are markedly and concurrently elevated in Iraqi patients whose respiratory *Candida albicans* infection was confirmed at the molecular level, and that each cytokine discriminated patients from healthy controls with complete accuracy. Interpreted together, these findings describe a mixed immunological state in which pro-inflammatory activation and Th2 polarisation proceed in parallel rather than in sequence, a pattern that is increasingly recognised as characteristic of fungal disease of the airway.

The isolation rate of 21.0% recorded in our respiratory cohort is consistent with the broader literature on *Candida* recovery from respiratory specimens. A ten-year retrospective analysis of lower respiratory secretions identified *Candida spp*. in 27.66% of non-contaminated samples, with *C. albicans* predominating(15), and a recent multicentre cohort of 11,925 patients hospitalised for acute exacerbation of chronic obstructive pulmonary

Table 3. Demographic Characteristics of patient and control groups.

Variable	Patients (n = 200) No.(%)/ Mean ± SD	Controls(n = 100) No.(%)/ Mean ± SD	*P*-value
Mean Age ± SD (years)	49.26 ± 18.25	53.36 ± 16.96	0.061, NS
< 40	71 (35.5%)	23 (23.0%)	
41 – 60	66 (33.0%)	41 (41.0%)	0.085 NS
≥ 60	63 (31.5%)	36 (36.0%)	
Male	94 (47.0%)	41(41.0%)	0.74 NS
Female	106 (53.0 %)	59 (59.0 %)	

NS: Non-significant($P > 0.05$). Independent t-test for mean age; Chi-square(χ^2) for categorical comparisons.

Table 4. *Candida albicans* culture and hwp1 PCR result among patients and controls.

Result	Patients (n = 200) No.(%)	Controls (n = 100) No.(%)	χ^2	*P* - value
Culture positive	42 (21.0%)	0 (0.0%)	22.70	< 0.0001****
Culture negative	158 (79.0%)	100 (100.0%)		
hwp1 PCR positive(941 bp)	37 (88.1% of positive*)	N/A	24.41	< 0.0001****
Hwp1 PCR negative	5 (11.9% of positive*)	N/A		

****$P \leq 0.0001$.Chi-square(χ^2) test.* Percentage calculated from 42 culture-positive isolates. N/A: PCR not applied to culture-negative controls.

Table 5. Serum TNF-α and IL-4 concentrations in confirmed *Candida* patients and healthy controls.

Parameter n = 142	Mean ± SD		*P*-value
	Healthy Control n = 100	Candidiasis Patients n = 42	
TNF-α concentration (ng/L)	34.85 ± 14.73	156.6 ± 38.20	< 0.0001****
IL-4 (ng/L)	17.78 ± 6.783	56.59 ± 17.19	< 0.0001****

****$P \leq 0.0001$. Independent samples t-test. TNF-α: Tumor necrosis factor-Alpha; IL-4: Interluekin-4; SD: Standard Deviation.

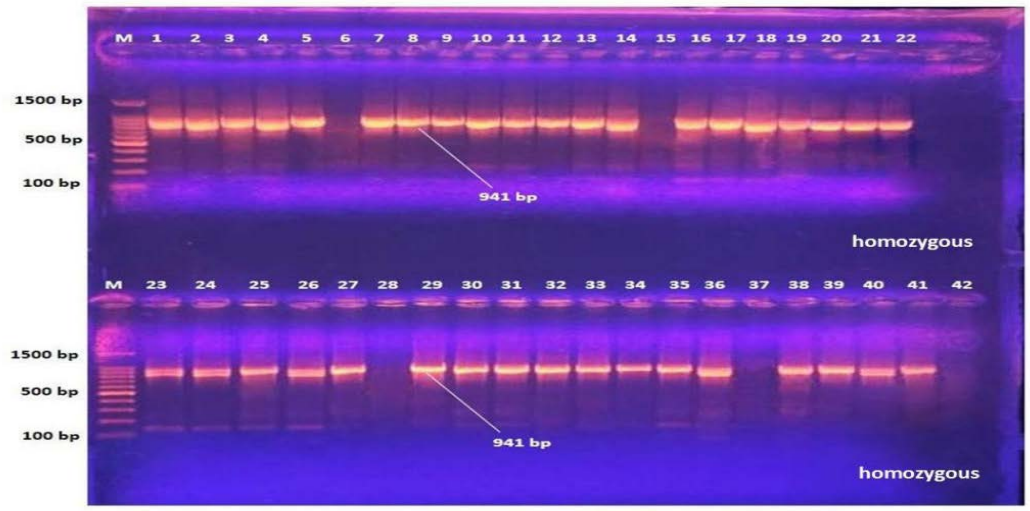

Figure 1. Agarose gel electrophoresis of hwp1 PCR products of the 42 confirmed *C. albicans* isolates. Upper panel: M = 1.5 kb DNA ladder; Lanes 1– 22. Lower panel: M = 1.5 kb DNA ladder; Lanes 1– 5, 7_14, 16_27, 29_ 36, 38_4 display a single band at 941 bp, consistent with the Class IV homozygous HWP1 genotype of *Candida albicans* (37/42; 88.1%). Lanes 6, 15, 28, 37 and 42 show no amplification product (hwp1 PCR-negative isolates).

Table 6. ROC Curve analysis: diagnostic Accuracy Serum TNF-α and IL-4.

Parameter	TNF-α	IL-4
AUC	1.000	1.000
95% Confidence Intervals	1.000 - 1.000	1.000 - 1.000
Optimal cut-off	>71.21 ng/L	>29.75 ng/L
Sensitivity	100.0%	100.0%
Specificity	100.0%	100.0%
P-value	<0.0001 ****	<0.0001 ****

AUC: Area Under the Curve. **** $P<0.0001$.

disease reported *Candida* positivity in 10.5% of cases(16). Reported prevalence varies widely with the underlying disease, ranging from 10–40% in stable chronic obstructive pulmonary disease to more than 90% during acute exacerbations, and from approximately 30% in non-cystic-fibrosis bronchiectasis to 40–60% in cystic fibrosis(17). Our figure therefore occupies the expected middle range for a mixed respiratory population, and the complete absence of *Candida* growth among healthy controls reinforces the pathological rather than incidental nature of these isolates.

Molecular confirmation added an important layer of specificity. The expected 941 bp hwp1 amplicon, corresponding to the homozygous *C. albicans* genotype, was obtained in 37 of 42 culture-positive isolates (88.1%). The five non-amplifying isolates most plausibly represent non-albicans species misassigned at the phenotypic stage. This interpretation is directly supported by contemporary work on hwp1 size polymorphism, in which amplicon length reliably separates *C. albicans* (941 bp) from *C. dubliniensis* (569 bp), *C. africana* (700 bp) and *C. stellatoidea* (800 bp), and in which a proportion of chromogenic-agar isolates presumptively identified as *C. albicans* were subsequently reassigned to other genera by proteomic and molecular methods(18). Such discordance underscores that culture and chromogenic identification, even when supplemented by automated systems, remain insufficient for definitive speciation and that hwp1 amplification provides a pragmatic and inexpensive confirmatory step.

The approximately 4.5-fold elevation of TNF-α observed in our patients is biologically coherent with the established position of this cytokine at the apex of innate antifungal defence. Recognition of β-glucan and mannan through Dectin-1 and Dectin-2 receptors drives macrophage and monocyte TNF-α release, which in turn sustains phagocyte activation, neutrophil recruitment and Th1 polarisation (8,19). The functional necessity of this axis is evidenced by the increased incidence of invasive candidiasis in patients receiving anti-TNF biologics or harbouring defects in TNF-α signalling(10). Our observation aligns closely with regional data: Mezher and colleagues documented simultaneous elevation of TNF-α and IL-22 in pulmonary candidiasis in Salah Al-Din province and attributed the response to Dectin-2-mediated macrophage activation(20), while a case-control study conducted in Nasiriyah reported significantly raised IL-1β, IL-6, TNF-α, IFN-γ and IL-10 alongside elevated C-reactive protein, procalcitonin and β-D-glucan in patients with invasive *C. albicans* infection(21). The present data from Wasit province extend this pattern to molecularly confirmed respiratory disease.

The concomitant rise in IL-4 is the more interpretively demanding observation. As the canonical Th2-inducing cytokine, IL-4 suppresses M1 macrophage polarisation, restrains pro-inflammatory cytokine secretion and diminishes the oxidative killing capacity required for effective fungal clearance(11,12). Sustained IL-4 signalling has been linked to defective cell-mediated antifungal immunity and to chronic mucocutaneous candidiasis in primary immunodeficiency(22). Recent evidence lends further mechanistic plausibility to a Th2 component in airway candidiasis: experimental gut dysbiosis with *C. albicans* overgrowth expands lung-resident type 2 innate lymphoid cells and amplifies Th2-driven airway inflammation, and higher intestinal fungal-to-bacterial

DNA ratios have been associated with more severe asthma exacerbations in human cohorts(17). The recognition of allergic bronchopulmonary candidiasis as a distinct hypersensitivity entity characterised by elevated total and *Candida*-specific IgE similarly situates IL-4 within the spectrum of *Candida*-associated pulmonary pathology(17). The elevation observed here therefore appears to reflect an immunological misdirection that coexists with, and partially counteracts, the TNF-α-driven effector response, thereby favouring fungal persistence. Contemporary reappraisals of *Candida* in the lung reach a convergent conclusion, arguing that the organism is not a passive contaminant but a modifier of host immune responses capable of exacerbating lung injury(17,23).

The absence of a significant association between the two cytokines ($r = 0.182$, $P = 0.249$) suggests independent cellular origins rather than a shared regulatory circuit, with TNF-α derived predominantly from Dectin-stimulated mononuclear phagocytes and IL-4 from mast cells, basophils or innate lymphoid populations responding through alternative pathways (3, 16). Of greater immediate clinical relevance is the inverse correlation between TNF-α and procalcitonin ($r = -0.339$, $P = 0.028$). Procalcitonin is induced principally by bacterial endotoxin and remains comparatively low in pure fungal infection(24); patients mounting a stronger fungal-specific response thus displayed a lesser concurrent bacterial inflammatory component. This complementarity may assist in the differentiation of fungal from bacterial respiratory disease, although procalcitonin also carries prognostic weight in *Candida*-positive respiratory and bloodstream infection(16,25), and *Candida* isolation from the airway has been associated with worse short-term outcomes and increased three-year mortality(16). Early recognition therefore retains clinical importance, particularly given the persistent attributable mortality of invasive candidiasis(26).

Finally, the area under the curve of 1.000 for both cytokines indicates complete separation of patient and control distributions at the derived cut-offs. While statistically striking, this result must be regarded cautiously: the modest number of confirmed cases ($n = 42$), the single-region design and the comparison against wholly healthy rather than disease-matched controls plausibly inflate apparent accuracy. Comparative work indicates that no single biomarker performs optimally in isolation and that multimarker panels offer superior diagnostic and prognostic discrimination(27-31). The consistency of our findings across three independent lines of evidence nonetheless supports the combined use of serum TNF-α and IL-4 with hwp1 PCR, pending validation in larger, multicentre cohorts that include patients with bacterial pneumonia and *Candida* colonisation as comparators.

Conclusion

Serum TNF-α and IL-4 were both substantially elevated in Iraqi patients with hwp1-confirmed*Candida albicans* respiratory infections, achieving perfect diagnostic accuracy at established optimal cut-off values. Their concurrent elevation captures a paradoxical immune state pro-inflammatory activation alongside Th2-driven suppression may indeed reflect a mixed or dysregulated immune response that may contribute to fungal persistence in the respiratory tract. The inverse correlation between TNF-α and procalcitonin further distinguishes the immunological signature of fungal from bacterial respiratory disease. Combined measurement of serum TNF-α and IL-4 alongside hwp1 PCR confirmation offers a diagnostically robust and clinically accessible approach to confirming *Candida albicans* respiratory infection in healthcare settings.

Acknowledgments

The authors would like to thank the clinical and laboratory staff of Al-Zahraa Teaching Hospital, Al-Karama Teaching Hospital, and the Respiratory Diseases Clinic in Al-Kut for their cooperation in sample collection. Special thanks to Department of Microbiology, College of Medicine, University of Thi-Qar.

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Competing Interests

None.

Data Availability Statement

Datasets that were used and analyzed in this current research are accessible to the author on reasonable requests.

Ethical Approval

The study was approved by the Institutional Research Ethics Committee (IREC) of the College of Medicine, University of Thi-Qar (Approval ID: IQ.UTQ.MED.2024.030, date: 17\11\2025). All participants provided informed consent.

Funding

None.

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