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Original Article

Bronchoalveolar lavage: A key tool in pulmonary fungal infection management

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ABSTRACT

Objective: Bronchoalveolar lavage (BAL) is a widely used diagnostic tool for evaluating lower respiratory tract infections and pulmonary pathologies in children. BAL provides valuable diagnostic information and can significantly influence management strategies, particularly in patients with malignancy, immunodeficiency (ID), or critical illness. This study aimed to assess the diagnostic and therapeutic value of BAL in pulmonary fungal infections.

Material and Methods: This retrospective study analyzed BAL samples obtained by flexible fiberoptic bronchoscopy (FFB) between 2019 and 2024 in the Pediatric Pulmonology Department of a tertiary referral center. Among 668 patients who underwent FFB, those with BAL fungal culture, BAL *Aspergillus* DNA PCR, and BAL galactomannan antigen (GM) testing were included. Demographic, clinical, radiological, and bronchoscopic findings, as well as antifungal treatment modifications based on BAL results, were evaluated.

Results: BAL results from 668 patients were reviewed. Fungal culture was performed in 236 (35.3%), *Aspergillus* DNA PCR in 127 (19%), and GM in 121 (18.1%) patients. Among those tested for fungal culture, 58.9% were male, with a mean age of 7.0 years (±5.7). Fungal culture positivity was found in 26 (11%) patients: *Candida albicans* (n=14), *Aspergillus spp.* (n=5), *Candida dubliniensis* (n=3), *Aspergillus* fumigatus (n=3), and *Penicillium spp.* (n=1). Culture positivity was significantly higher in patients with malignancy (p=0.011). BAL *Aspergillus* DNA PCR was positive in two patients, and BAL GM in five. Based on BAL results, antifungal therapy was modified in seven patients.

Conclusion: BAL fungal culture, BAL GM, and *Aspergillus* PCR testing are valuable diagnostic tools for pulmonary fungal infections in children, especially those with malignancy or immunodeficiency (ID). The observed treatment modifications following BAL results emphasize their clinical importance not only in establishing diagnosis but also in guiding and optimizing antifungal therapy.

Keywords: *Aspergillus, candida,* bronchoalveolar lavages, bronchoscopy, pulmonary fungal infections

Introduction

Bronchoalveolar lavage (BAL) is a commonly used and relatively safe diagnostic procedure for evaluating patients with lung disease. When conventional assessments such as clinical history, physical examination, laboratory tests, pulmonary function testing, and imaging are inconclusive, BAL provides valuable diagnostic information (1-3). In children with underlying conditions such as ID, critical illness, or malignancy—where the incidence of opportunistic infections is increased—BAL plays a crucial role in guiding management strategies (4-6).

Bronchoalveolar lavage provides valuable etiological information in suspected pulmonary infections when noninvasive diagnostic methods are inconclusive. Microbiological analysis of bronchoalveolar lavage fluid supports targeted antimicrobial therapy and informs clinical management (7-9). Fungal infections represent a significant proportion of pulmonary infections in high-risk patients, and

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bronchoalveolar lavage fluid analysis plays an important role in their diagnosis (10).

Fungal infections are becoming increasingly prevalent, particularly among immunocompromised patients and those with malignancies. Despite the availability of new drugs, mortality rates from fungal infections remain high in these patient groups (11). In various population-based studies, the positivity rate of fungal cultures from BALF obtained through FFB has been reported to range from 16% to 73% (12,13). In these studies, Candida species have been consistently reported as the most commonly isolated organisms in patients with positive fungal cultures from BALF (5,6). Immunocompromised patients with candidemia have a 40% mortality rate. The mortality rate attributable to invasive aspergillosis ranges from 30% to 70% (14,15).

Although fungal culture samples are sent from BALF for diagnosis of fungal infections, the newly developed polymerase chain reaction (PCR) method has provided a powerful tool to improve the detection and identification of fungal pathogens in clinical microbiology practice (16). The galactomannan antigen (GM) assay is essential for diagnosing invasive fungal infections, particularly in immunocompromised patients and those with hematological malignancies (14).

This study aimed to highlight the role of BALF in the diagnosis and assessment of pulmonary fungal infections.

Materials and Methods

This study retrospectively evaluated BALF results obtained via FFB in the Pediatric Pulmonology Ankara Bilkent City Hospital between January 2019 and December 2024. The BALF results of 668 patients who underwent FFB during this period were analyzed. The study included those for whom BAL fungal culture, BAL *Aspergillus* DNA PCR, and BAL GM tests were performed. The demographic data, clinical findings, laboratory results, radiological examination results, treatment, and follow-up of these patients were examined using the hospital's digital record system. The indications for FFB were evaluated and grouped according to etiology. The patients' BAL fungal culture, BAL *Aspergillus* DNA PCR, and BAL GM results, as well as the antifungal treatments or prophylaxis they received, were evaluated. All bronchoscopy procedures and clinical decisions regarding the indication for bronchoscopy were made and performed by the same experienced team throughout the study period in order to ensure consistency. In patients with significant underlying comorbidities such as ID or malignancy, prolonged hospitalization, or strong clinical suspicion of fungal infection, additional tests including fungal culture, *Aspergillus* PCR, and GM assay were requested from BAL fluid. Furthermore, in patients hospitalized due to malignancy who had positive serum GM or serum *Aspergillus* PCR results, BAL *Aspergillus* PCR and BAL GM testing were also specifically performed.

Informed consent for bronchoscopy procedure was obtained from the parents of all patients. Prior to the procedure, informed preoperative anesthesia consent was obtained and a fasting period of at least eight hours was ensured. The procedure was performed under operating room conditions. General anesthesia was administered by an anesthesiologist, and ventilation was maintained using a laryngeal mask airway. A fiberoptic bronchoscope appropriate for the patient's age and body weight was selected and advanced through the laryngeal mask airway to perform the FFB.

Statistical analysis

Statistical analysis were performed using the IBM Statistical Package, version 26.0 (SPSS Inc., Armonk, NY, IBM Corp., USA). Numerical data with a normal distribution were expressed as the mean and standard deviation. Percentages (%) and numbers (n) were used to express categorical variables. Categorical variables were analyzed using the Chi-Square test. Age was analyzed using the Independent Samples t-test. In all statistical tests, $p < 0.050$ was considered statistically significant.

Results

Patient characteristics and sample overview

BAL results from 668 patients who underwent FFB during the study period were evaluated. BAL fungal culture was requested for 236 (35.3%), BAL *Aspergillus* DNA PCR for 127 (19%), and BAL GM for 121 (18.1%) patients. Of the 236 patients tested for fungal culture, 139 (58.9%) were male and 97 (41.1%) were female, with a mean age of 7.0±5.7 years. The mean ages of female and male patients were 7.6±5.9 years and 6.5±5.2 years, respectively, with no significant age difference between the genders (p=0.960).

FFB indications and underlying diseases

The indications for FFB and the underlying diagnoses of the 236 patients who underwent BAL for fungal culture were analyzed. Among these, 35 (14.8%) patients had ID, 22 (9.3%) had malignancy, and 56 (23.7%) presented with atelectasis. A history of lower respiratory tract infection (LRTI) was documented in 78 (33.0%) patients, and 33 (13.9%) were evaluated for suspected interstitial lung disease or bronchiolitis obliterans (ILD/BO). FFB was also performed in six (3.2%) patients with a history of tracheoesophageal fistula (TEF), in three (1.6%) patients due to extubation intolerance, and in three (1.6%) patients presenting with stridor.

Radiological imaging findings of patients undergoing FFB

Thoracic computed tomography (CT) was available for 197 out of the 236 patients who underwent BAL fungal culture testing. Radiological findings included fibroatelectatic changes (n=76), ground-glass opacities (n=61), bronchiectasis (n=57), consolidation (n=29), nodular infiltrates (n=18), mosaic perfusion pattern (n=9), pleural

effusion (n=6), and mass-like lesions (n=5). Several patients demonstrated multiple radiological findings. CT scans were reported as normal in seven (2.9%) patients.

FFB findings of patients undergoing FFB

The FFB findings of the 236 patients who underwent BAL fungal culture were classified. Purulent or seropurulent secretions were observed in 115, tracheomalacia/bronchomalacia in 31, anatomical airway stenosis in 28, increased vascularity in 12, postoperative findings related to TEF in six, mucosal pallor in five, and granulation tissue in four patients. Other, less common findings (seen in a total of 18 patients) included epiglottic edema, tracheal mucosal folds, tracheal dyskinesia, hemorrhage, subglottic stenosis, polypoid structures, mucosal lesions, pulsatile masses, white mucosal plaques, and fibrinoid tissue.

Evaluation of fungal culture results according to ffb indications, ct findings, and FFB findings

Fungal culture positivity was detected in 26 of the 236 patients for whom BAL fungal culture testing was requested (11%). *Candida albicans* was identified in 14 patients, *Aspergillus* species in five, Candida dubliniensis in three, *Aspergillus* fumigatus in three patients, and Penicillium species in one patient.

Among these 26 patients, malignancy was the most frequent underlying condition (six patients, 23%), followed by atelectasis and LRTI (five patients each, 19.3%). ID and bronchiectasis were present in three patients each (11.6%), and hemoptysis in two patients (7.6%). Tuberculosis and sarcoidosis were each diagnosed in one patient (3.8%). In patients who underwent FFB due to malignancy, BAL fungal culture positivity was more frequent compared to other indications (72.7% vs. 27.3%, p=0.011). There was no significant difference in the distribution of cultured fungal pathogens between immunodeficient patients and those with malignancies. BAL fungal culture positivity according to FFB indications and underlying diseases are summarized in Table I.

Fungal culture positivity varied according to thoracic CT findings: 55.5% (10/18) in patients with nodular infiltration, 33.3% (2/6) with pleural effusion, 8.7% (5/57) with bronchiectasis, 11.4% (7/61) with ground-glass opacities, and 7.9% (6/76) with atelectasis. Patients were then grouped based on the presence or absence of nodular infiltration and/or ground-glass opacities. BAL fungal culture positivity was significantly higher in patients with these findings compared to those without (60.9% vs. 39.1%, p <0.010).

Table I: BAL fungal culture positivity according to FFB indications and underlying diseases

Bronchoscopy indications	BAL Fungal Culture		p†
	Positive	Negative	
Malignancy*	6 (27.3)	16 (72.7)	0.011
ID*	3 (8.6)	32 (91.4)	0.617
Atelectasis*	3 (5.4)	53 (94.6)	0.147
Recurrent LRTI*	10 (12.8)	68 (87.2)	0.517
Suspected ILD/BO*	3 (9.1)	30 (90.9)	0.703

:** *n(%),* **†:** *Chi-Square test,* ***LRTI: *Lower Respiratory Tract Infection,* ***ILD:*** *Interstitial Lung Disease,* ***BO:*** *Bronchiolitis Obliterans,* ***ID:*** *Immunodeficiency*

Table II: BAL fungal culture positivity according to FFB findings

Bronchoscopic findings	BAL fungal culture		p‡
	Positive	Negative	
Purulent or seropurulent secretions*	20 (17.4)	95 (82.6)	0.002
Tracheomalacia/bronchomalacia*	2 (6.5)	29 (93.5)	0.384
Anatomical airway stenosis*	3 (10.7)	25 (89.3)	0.957
Mucosal pallor*	0 (0.0)	5 (100.0)	0.426
Postoperative findings related to TEF*	0 (0.0)	6 (100.0)	0.383
Granulation tissue*	1 (25.0)	3 (75.0)	0.368
Increased vascularity*	0 (0.0)	12 (100.0)	0.211
Other*†	0 (0.0)	18 (100.0)	0.120

:** *n(%),* **†:** *Epiglottic edema, tracheal mucosal folds, tracheal dyskinesia, hemorrhage, subglottic stenosis, polypoid structures, mucosal lesions, pulsatile masses, white mucosal plaques, fibrinoid tissue etc,* **‡:** *Chi-Square test,* ***TEF: *tracheoesophageal fistula*

BAL fungal culture positivity was evaluated according to FFB findings. Patients in whom purulent or seropurulent secretions were observed on FFB had a higher frequency of positive fungal cultures compared to those with other findings (82.6% vs. 17.4%, p=0.002). BAL fungal culture positivity according to FFB findings is summarized in Table II.

Antifungal prophylaxis and treatment use at the time of FFB

Among patients for whom BAL fungal cultures were requested, 21 (8.9%) were receiving antifungal prophylaxis at the time of FFB (10 on fluconazole, seven on voriconazole, and four on itraconazole). Of these, 13 had ID and eight had malignancy. BAL fungal cultures were negative in 17 patients receiving prophylaxis. In four patients receiving prophylaxis, BAL fungal culture positivity was detected; therefore, prophylaxis was discontinued, and a new antifungal treatment was initiated.

Additionally, 25 (10.5%) patients were receiving antifungal treatment at the time of FFB. Among these, 23 (92%) had negative fungal cultures. Antifungal treatment decisions were made collaboratively with the pediatric infectious diseases team based on clinical status, infection markers, and lung imaging. Antifungal treatment was modified in two patients due to positive fungal cultures with *Aspergillus* species, specifically *Aspergillus* fumigatus. Out of 26 patients with positive fungal culture, three (11%) died; two of them had ID, and one had an underlying malignancy.

BAL *Aspergillus* DNA PCR and BAL galactomannan antigen test results

BAL *Aspergillus* DNA PCR was positive in two patients. One patient, diagnosed with acute lymphoblastic leukemia and not on antifungal treatment or prophylaxis, started voriconazole after the positive PCR result; this patient also had fungal culture positivity. The other patient, diagnosed with chronic granulomatous disease analysis and not receiving antifungal therapy, was started on voriconazole following PCR positivity despite a negative fungal culture. Both patients had purulent or seropurulent secretions observed during FFB and nodular infiltrates on thoracic CT. Their BAL GM tests were also positive.

In addition to these two patients, three more had positive BAL GM results. One patient with ID was started on amphotericin B after the positive outcome. Another patient with malignancy and fungal culture positivity was receiving voriconazole at the time of FFB; treatment was not changed due to clinical stability. The third patient, also with malignancy and on voriconazole, continued treatment as clinical status remained stable.

Discussion

In our study, we demonstrate that BAL findings significantly influenced treatment decisions in patients evaluated for pulmonary fungal infections. Notably, BAL fungal culture positivity was significantly more frequent in patients undergoing FFB for malignancy than in other patient populations. Evaluation of culture positivity based on FFB findings revealed that patients with observed purulent or seropurulent secretions had a significantly higher rate of BAL fungal culture positivity. Fungal culture positivity was more frequent in patients presenting with nodular infiltration and/or ground-glass opacities on thoracic CT. Despite receiving antifungal prophylaxis, some patients required modifications in antifungal therapy due to positive BAL fungal culture results.

A study involving 35 pediatric patients in the intensive care unit (ICU) evaluated BAL fungal cultures, *Aspergillus* PCR, GM and Candida mannan antigen. The majority of patients had malignancies and had been hospitalized in the ICU for more than one week. Chest radiography was the primary imaging modality, with patchy consolidations and nodular lesions being the most common findings (17). BAL fungal cultures were positive in 15 patients: *Candida spp*. in 12, *Aspergillus spp*. in one, and both in two patients (17). In our study, similar to the referenced study, culture positivity was more frequent in patients with malignancy, and *Candida spp*. was the most commonly isolated organism. However, the BAL culture positivity rate in our study was slightly lower than that reported in other studies (12,13,17). This difference may be attributed to variations in patient populations, as our cohort included not only hospitalized patients but also clinically stable outpatients who underwent FFB. Also, some patients who underwent FFB may have had no detectable pathogens due to antifungal therapy at the time of the procedure.

In a study involving 101 patients with malignancy who were evaluated for suspected pulmonary fungal infection, 24 patients underwent BAL, 31 underwent biopsy (27 lung, 4 sinus), and 46 were assessed by thoracic CT for diagnostic purposes (18). Among the 38 patients with thoracic CT findings suggestive of fungal infection, antifungal treatment was modified in 30 cases. Treatment changes were also made in two of the six patients with positive BAL fungal cultures and in four of the 12 patients with biopsy-confirmed fungal infection (19). In another study conducted on adult patients, fungal growth was detected in 16% of cases; antifungal therapy was initiated in 15% of these patients, and treatment was modified in 7% (13). Similar to the referenced studies, our cohort also exhibited changes in antifungal treatment in a total of seven (26%) patients due to BAL results. These changes demonstrate the impact of BAL results on the regulation of treatment.

In a study conducted on adult solid organ transplant recipients, 131 BAL samples were evaluated. BAL fungal cultures were positive in 24 patients, and BAL *Aspergillus* PCR was positive in nine patients (13). None of the BAL GM tests were positive. Based on clinical and radiological findings, BAL *Aspergillus* PCR demonstrated moderate sensitivity and high specificity for the diagnosis of fungal infections (13). In another study, BAL samples obtained from 1736 FFB procedures in 688 ICU patients diagnosed with pneumonia were evaluated (20). *Aspergillus spp* was identified in 18 of 968 BAL fungal cultures, and 53 of 1146 BAL GM samples tested positive. Due to technical limitations, *Aspergillus* PCR was not performed (20). While the BAL GM positivity rate in that study was comparable to that observed in our cohort, the proportion of patients with *Aspergillus spp* in BAL fungal cultures was slightly higher in our center (3.4% vs. 1.9%). This difference may be attributed to variations in patient selection and the underlying burden of comorbidities. The findings highlight the importance of not only BAL fungal culture but also BAL GM and BAL *Aspergillus* PCR tests in supporting the diagnosis of suspected pulmonary fungal infections.

Our findings support the diagnostic value of BAL samples in the assessment of pulmonary fungal infections. BAL fungal culture, BAL GM, and BAL *Aspergillus* PCR tests are particularly relevant in managing patients with malignancy or ID. Treatment adjustments based on BAL findings appear to improve clinical outcomes. However, large-scale, longitudinal studies with comprehensive data are required to validate these observations.

Ethics committee approval

This study was conducted in accordance with the Helsinki Declaration Principles. The study was approved by Bilkent City Hospital (25.06.2025, reference number: TABED-2-25-1337).

Contribution of the authors

Study conception and design: HY, GDT, DAT, SEP, GC; data collection: HY, ÇY, SÖT; analysis and interpretation of results: HY, ŞSAS, IB, MKÇ; draft manuscript preparation: HY, BET, GAD, AÜ. All authors reviewed the results and approved the final version of the article.

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Conflict of interest

The authors declare that there is no conflict of interest.

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