Master of Medicine

Diagnostic yield of a bronchoalveolar lavage fluid galactomannan assay in patients with negative serum galactomannan results suspected to have invasive pulmonary aspergillosis

혈청 갈락토만난이 음성으로 확인된
침습성 폐 아스페르길루스증 의심환자에서
기관지 폐포 세척액 갈락토만난의 진단적 유용성

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Diagnostic yield of a bronchoalveolar lavage fluid galactomannan assay in patients with negative serum galactomannan results suspected to have invasive pulmonary aspergillosis

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Abstract

Background: There are limited data in real clinical practice on the diagnostic value of a bronchoalveolar lavage (BAL) fluid galactomannan (GM) assay in patients with suspected invasive pulmonary aspergillosis (IPA) who had negative serum GM results. Thus, we investigated the diagnostic performance of a BAL GM assay in patients with negative serum GM assay results who were suspected to have IPA.

Methods: This study was performed between May 2008 and April 2019 at a tertiary care hospital in Seoul, South Korea. All patients with suspected IPA whose serum GM assays revealed negative results who sequentially underwent BAL were enrolled in this study. Patients were classified as having disease that was proven, probable, possible and not IPA the revised definition of European Organization for Research and Treatment of Cancer and the Mycoses Study Group in 2019.

Results: A total of 341 patients with suspected IPA including 4 cases of proven IPA, 38 case of probable IPA, 107 cases of possible IPA, and 192 patients without IPA were enrolled. Of these 341 patients, 107 (31%) with possible IPA were excluded from the final analysis. Of 42 patients with proven and probable IPA who had initial negative serum GM results, 24 (57%) had positive BAL GM results (n = 24) or BAL fungal culture results (n = 8). Among the remaining 18 (43%) patients, 2 (5%) were diagnosed with proven IPA by histopathologic examination of a transbronchial lung biopsy, 6 (14%) with probable IPA by a subsequent sputum fungal culture, and 12 (29%) with probable IPA by a repeated serum GM assay after BAL. Of 192 patients without IPA, 14 (7%) had positive BAL GM results (n = 14) or BAL fungal culture results (n = 8). In addition, BAL revealed evidence of other opportunistic infections including *Pnuemocystis. jirovecii* pneumonia (14% [26/190]), cytomegalovirus (CMV) pneumonia (5% [9/188]), and respiratory viral pneumonia (6% [12/193]).

Conclusion: BAL in patients with suspected IPA who had initial negative serum GM results provided additional diagnostic yield in approximately half of patients with the evidence of another co-infection.

Keywords: Invasive Pulmonary Aspergillosis, Bronchoalveolar Lavage, Galactomannan

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INTRODICTION

Invasive pulmonary aspergillosis (IPA) is a common and fatal opportunistic infection in patients with prolonged neutropenia, hematologic malignancy and hematologic stem cell transplantation; it is caused by filamentous fungi of the genus Aspergillus [1]. Invasive aspergillosis is characterized by progression of the infection across tissues but performing a lung biopsy to confirm a histopathological diagnosis of proven IPA is difficult in immunocompromised patients. Since the sensitivity of a mycologic culture is as low as 20% [2], other mycologic evidence such as serum galactomannan (GM) testing has been widely used for the diagnosis of IPA.

However, the sensitivity of a serum GM assay is still suboptimal at 65% with a cutoff value of 1.0 [3]. A previous study reported that the sensitivity of a bronchoalveolar lavage (BAL) fluid GM assay is 91.3% at a cutoff value of 1.0 [4]. Therefore, BAL has been considered a sequential diagnostic procedure in patients with negative serum GM results but with disease suspicious of IPA because of the relative invasiveness of the procedure [5]. However, there are limited data in real clinical practice on the diagnostic value of performing subsequent BAL GM assays in patients with suspected IPA with negative serum GM results. Thus, we investigated the diagnostic performance of a BAL GM assay in patients with negative serum GM assay results who were suspected to have IPA.

METHODS

Study population and patient selection

A retrospective study was conducted involving patients who were admitted to the hematology unit at a 2700 bed tertiary-care teaching hospital in Seoul, South Korea, and underwent subsequent BAL for suspected IPA between May 2008 and April 2019. The electronic medical records of patients with suspected IPA were reviewed, and patients who had positive serum GM results before BAL were excluded. In the case of patients who underwent BAL more than once, only the GM assay from the first BAL was analyzed. This study was approved by the institutional review board in our hospital.

Definitions

Patients were classified as having disease that was proven, probable, possible and not IPA the revised definition of European Organization for Research and Treatment of Cancer and the Mycoses Study Group in 2019 [6]. Proven IPA was defined as histopathological evidence of tissue invasion of hyphae morphologically consistent with Aspergillus. Probable IPA was defined as the presence of host factors with clinical features such as dense, well-circumscribed lesions with or without a halo sign, an air crescent sign, or a cavity and wedge-shaped and segmental or lobar consolidation on CT, and mycological evidence of fungal infection by a culture or GM assay.

Neutropenia was defined as recent history of neutropenia (<500 neutrophils/mm3 for >10 days) related to a present infection. Steroid use was defined as ≥0.3 mg/kg of methylprednisolone for ≥3 weeks in the past 60 days. Immunosuppressant use was defined as treatment with T-cell or B-cell immunosuppressants during the past 90 days.

Mycological and other microbiologic evaluation

The Platelia Aspergillus EIA (Bio-Rad Laboratories, Inc., Hercules, California, USA) was used to detect the presence of GM from serum and BAL samples. A BAL GM result larger than 10 was calculated as 10 because the exact value over 10 could not be detected with this assay. The cutoff value of the serum GM and BAL GM assay for the diagnosis of IPA was 1.0 with the combination of a serum GM value more than 0.7 and a BAL GM value more than 0.8, according to 2019 EORTC-MSG criteria [5].

Other (opportunistic) infections that were identified from BAL were defined as clinically significant pathogens identified only from BAL fluid other than a nasopharyngeal swab/aspirate or sputum culture: Pneumocystis jirovecii, cytomegalovirus (CMV), and respiratory virus. P. jirovecii pneumonia (PCP) was defined as positive immunohistochemical staining or a cycle threshold (Ct) value of quantitative PCR <31 of BAL fluid [7] with clinical and radiological evidence of pneumonia.

The diagnosis of a probable CMV pneumonia was performed through the detection of CMV in culture of BAL fluid or quantitation of CMV DNA in BAL fluid with clinical symptoms and/or signs of pneumonia [8]. Although a definitive cutoff for a CMV DNA load to differentiate pneumonia from pulmonary shedding does not exist, a quantitative PCR titer of CMV DNA >3.9 log IU/mL from BAL fluid was used to differentiate CMV pneumonia [9]. Probable or possible respiratory viral pneumonia was defined by the detection of respiratory viruses by viral culture without concomitant infection with other pathogens or multiplex reverse-transcription PCR using Seeplex RV15 ACE Detection (Seegene Inc., Seoul, Republic of Korea) from BAL samples only, respectively, as applied in our previous study [10].

Statistical analysis

Statistical analysis was performed with SPSS Statistics for Windows, version 24.0 (IBM Corp., Armonk, NY, USA). We used the Chi-squared test or Fisher's exact test as appropriate for the analysis of categorical variables and Student's t-test or Mann-Whitney U test as appropriate for continuous variables. The sensitivity, specificity, positive predictive value, negative predictive value, and likelihood ratios were calculated for the BAL GM assay with 95% confidential intervals. The cutoff value of the BAL GM assay derived from a receiver operating characteristic (ROC) curve was defined according to Youden's index [11]. All tests of significance were two-tailed, and a significance level of 0.05 was employed for all analyses.

RESULTS

Baseline clinical characteristics

The schematic flow chart of patient diagnosis is shown in Figure 1. A total of 585 patients who were admitted to the hematology unit and underwent bronchoscopy for suspected IPA over the period between May 2008 and April 2019 were reviewed. A total of 341 patients who had classical host factors for IPA and negative serum GM results before BAL were enrolled in this study. Of these 341 patients suspected with IPA, we excluded 107 (31%) with possible IPA, and the remaining 234 (69%) patients with 4 proven IPA, 38 with probable IPA, and 192 without IPA were included for the final analysis. Of 42 patients with proven and probable IPA who had initial negative serum GM results, 24 (57%) had positive BAL GM results (n = 24) or BAL fungal culture results (n = 8). Among the remaining 18 (43%), 2 (5%) were diagnosed with proven IPA by a histopathologic examination of a transbronchial lung biopsy, 6 (14%) were diagnosed with probable IPA by a subsequent sputum fungal culture, and 12 (29%) were diagnosed with probable IPA by a repeated serum GM assay after BAL.

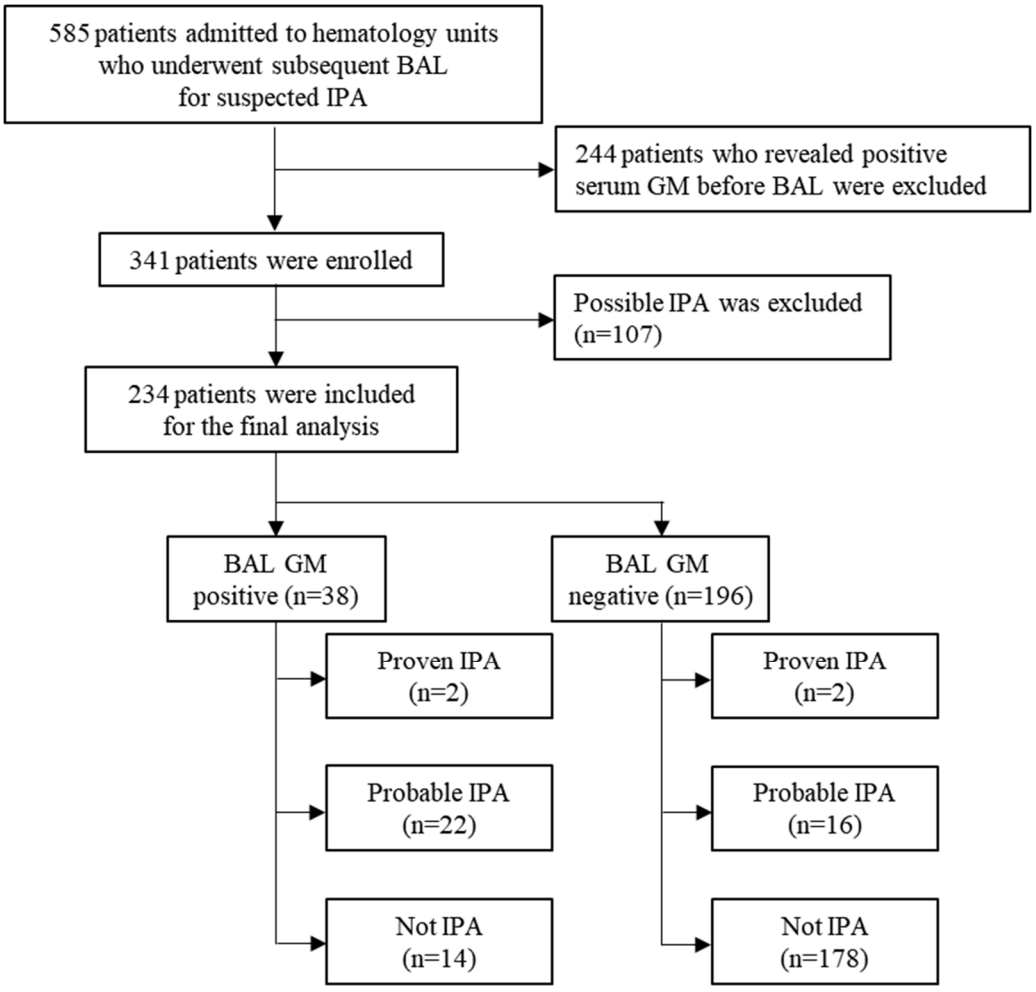


Figure 1. Flow chart of the diagnosis of patients included in this study.

The baseline clinical characteristics are presented in Table 1. There was no significant difference in characteristics between proven or probable IPA and not IPA except the full-matched allogenic hematopoietic stem cell transportation (HSCT) (p value = 0.01).

Table 1. Clinical and hematologic characteristics of the study population included in the final analysis

Characteristics	Total (n = 234)	Proven and Probable IPA (n = 42)	Not IPA (n = 192)	*p* value
Age, median years (IQR)	55 (45-64)	61 (52-66)	54 (44-63)	0.10
Female gender	88 (38)	21 (50)	67 (35)	0.07
Neutropenia (ANC<500/m^3)	65 (28)	15 (36)	50 (26)	0.21
Steroid[a] use	22 (9.4)	6 (14)	16 (8.3)	0.23
Median dose (mg) per kilogram (IQR)	0.70 (0.40-0.95)	1.50 (1.25-1.75)	0.70 (0.40-0.85)	0.94
Immunosuppressant[b] use	53 (23)	8 (19)	45 (23)	0.54
GVHD	49/86 (57)	10/15 (67)	39/71 (55)	0.40
Type of GVHD				
Acute	26/86 (30)	4/15 (27)	22/71 (31)	1.0
Chronic	23/86 (27)	6/15 (40)	17/71 (24)	0.21
Underlying diseases				
Myelodysplastic syndrome	34 (15)	6 (14)	28 (15)	0.96
Acute myeloid leukemia	101 (43)	18 (43)	82 (43)	0.76
Acute lymphoblastic leukemia	33 (14)	4 (10)	29 (15)	0.35
Myeloproliferative neoplasm	9 (4)	1 (2)	8 (4)	1.0
Lymphoma	32 (14)	7 (17)	25 (13)	0.53
Multiple myeloma	11 (5)	2 (5)	9 (5)	1.0

Others[c]	14 (6)	3 (7)	11 (6)	0.72
Allogenic HSCT	86 (37)	15 (36)	71 (37)	0.88
Type of HSCT				
Autologous	6/92 (7)	3/18 (17)	3/74 (4)	0.09
Allogenic, full-matched[d]	45/92 (49)	4/18 (22)	41/74 (55)	0.01
Allogenic, half-matched[e]	41/92 (45)	11/18 (61)	30/74 (41)	0.12
Mold-active antifungal agents before BAL	121 (52)	23 (55)	98 (51)	0.66
Median days from antifungal agents to BAL (IQR)	7 (2.5-18)	9 (4.0-16)	5.5 (2.0-19)	0.21
Amphotericin B	73 (31)	13 (31)	60 (31)	0.74
Caspofungin	11 (5)	2 (5)	9 (5)	1.0
Micafungin	3 (1)	0 (0)	3 (2)	1.0
Itraconazole	7 (3)	1 (2)	6 (3)	0.36
Voriconazole	18 (8)	4 (10)	14 (7)	0.54
Posaconazole	9 (4)	3 (7)	6 (3)	0.21
Other (opportunistic) infections identified by BAL				
Pneumocystis jirovecii	26/190 (14)	5/30 (17)	21/160 (13)	0.60
Cytomegalovirus	9/188 (5)	2/29 (7)	7/159 (4)	0.63
Respiratory virus	12/193 (6)	1/29 (0)	11/164 (7)	0.22
Adenovirus	2 (17)	0 (0)	2 (18)	
Metapneumovirus	1 (8.0)	0 (0)	1 (9)	
Parainfluenza virus	7 (58)	1 (100)	6 (55)	

Respiratory syncytial virus	2 (17)	0 (0)	2 (18)
Rhinovirus	2 (17)	0 (0)	2 (18)

Data are given as a number (percentage) of patients unless otherwise indicated.

Abbreviation: IQR, interquartile range; GVHD, graft-versus-host disease; HSCT, hematopoietic stem cell transplantation

aPeak dose of steroids used in recent 2 weeks

bIncluded azathioprine, cyclosporine, mycophenolate mofetil, and tacrolimus

cIncluded aplastic anemia, autoimmune neutropenia, hemophagocytic lymphohistiocytosis, idiopathic thrombocytopenic purpura, and Langerhans cell histiocytosis

d22 from a sibling and 23 from an unrelated donor

e14 from a child, 7 from a parent, 14 from a sibling, and 6 from an unrelated donor

Diagnostic performances of various tests

The diagnostic performances of various tests are presented in Table 2. Of 42 patients with proven and probable IPA who had initial negative serum GM results, 24 (57%) had positive BAL GM results. False-positive results were documented in 14 patients among 192 patients without IPA. Therefore, in patients suspected with IPA who had negative serum GM results, the sensitivity and the specificity of the BAL GM assay at a cutoff value of 1.0 for the diagnosis of proven and probable IPA was 57% (95% CI, 42-72%) and 93% (95% CI, 89-96%), respectively. The prevalence of IPA with negative serum GM results in our study was approximately 18%, and the negative predictive value and positive predictive value of the BAL GM assay were 91% and 63%, respectively (Table 2).

Table 2. Diagnostic performance of various diagnostic tests in patients with suspected IPA who had negative serum GM results

Proven and Probable IPA vs Not IPA	Sensitivity % (n/N [a], 95% CI)	Specificity % (n/N [b],95% CI)	PPV % (95% CI)	NPV % (95% CI)	Positive Likelihood ratio (95% CI)	Negative Likelihood ratio (95% CI)
BAL GM or BAL fungal culture	57 (24/42, 41–72)	93 (178/192, 89-96)	63 (49–75)	91 (88–94)	8.4 (4.8–15)	0.46 (0.32–0.65)
BAL GM	57 (24/42, 41–72)	93 (178/192, 89-96)	63 (49–75)	91 (88–94)	8.4 (4.8–15)	0.46 (0.32–0.65)
Subsequent repeated serum GM	29 (12/42, 16–45)	93 (178/192, 88-96)	46 (30-63)	86 (83–88)	3.9 (2.0–7.9)	0.77 (0.63–0.94)
Sputum fungal culture[c]	21 (9/42, 10-37)	97 (187/192, 94-99)	64 (39–84)	86 (83-87)	8.2 (2.9-23)	0.81 (0.69-0.95)
Tissue biopsy[d]	40 (4/10, 9.6–70)	100 (11/11, 100-100)	100 (100-100)	65 (42–87)	Not applicable	0.6 (0.36–1.0)

Abbreviation: CI, confidence interval; PPV, positive predictive value; NPV, negative predictive value

aNumber of patients with a positive test result/number of patients tested among those diagnosed with proven and probable IPA

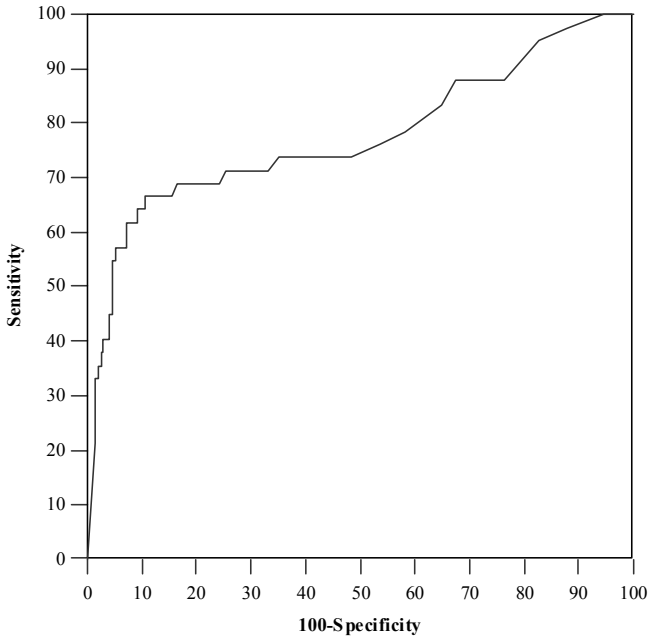
bNumber of patients with a negative test result/number of patients tested among those diagnosed with not IPA

C4 A. fumigatus, 5 unknown Aspergillus species

dIncluded transbronchial lung biopsy (n=3) and lobectomy (n=1)

Among the 42 patients with proven and probable IPA, the diagnoses of 12 (29%) patients were changed from possible IPA to probable IPA after subsequent repeated serum GM assays. Therefore, the sensitivity of subsequent repeated serum GM assays at a cutoff value of 1.0 for the diagnosis of proven and probable IPA in patients with suspected IPA who had negative serum GM results was 29% (95% CI, 21-50%).

The ability of the BAL GM assay to discriminate IPA as assessed by the area under the ROC curve as shown in Figure 2 was 0.78 (95% CI, 0.69-0.87). From the ROC curve of the BAL GM results, the optimal cutoff value was 0.69 by Youden's index with a sensitivity of 67% (95% CI, 55-84%) and a specificity of 89% (95% CI, 64-78%). The sensitivity of the BAL GM assay was 67% (95% CI, 50-80%), and the specificity was 88% (95% CI, 82-92%) at a cutoff value of 0.5, while the sensitivity was 48% (95% CI, 32-64%), and the specificity was 95% (95% CI, 91-98%) at a cutoff value of 1.5 (Supplemental Table 1).



BAL GM	
Area under the ROC curve	0.78 (95% CI, 0.69-0.87)
Cut-off value by Youden Index	0.69
Sensitivity	67% (95% CI, 55-84%)
Specificity	89% (95% CI, 64-78%)

Figure 2. ROC curve for BAL GM assay in patients suspected with IPA who had negative serum GM results.

Table 3. Diagnostic performance of the BAL GM assay by cutoff values in patients with suspected IPA who had negative serum GM results

Proven and Probable IPA vs Not IPA	Sensitivity % (n/N [a], 95% CI)	Specificity % (n/N [b],95% CI)	PPV % (95% CI)	NPV % (95% CI)	Positive Likelihood ratio (95% CI)	Negative Likelihood ratio (95% CI)
cut-off value						
≥ 0.5	67 (28/42, 50-80)	88 (168/192, 82-92)	54 (43-64)	92 (89-95)	5.3 (3.5-8.2)	0.38 (0.25-0.59)
≥ 0.69[c]	67 (28/42, 51-80)	89 (171/192, 84-93)	57 (46-68)	92 (89-95)	6.1 (3.9-9.6)	0.4 (0.24-0.58)
≥ 1.0	57 (24/42, 41–72)	93 (178/192, 89-96)	63 (49–75)	91 (88–94)	8.4 (4.8–15)	0.46 (0.32–0.65)
≥ 1.5	48 (20/42, 32-64)	95 (183/192, 91-98)	69 (52-82)	89 (86-92)	10 (5.0-21)	0.55 (0.41-0.73)

Abbreviation: CI, confidence interval; PPV, positive predictive value; NPV, negative predictive value

aNumber of patients with a positive test result/number of patients tested among those diagnosed with proven and probable IPA

bNumber of patients with a negative test result/number of patients tested among those diagnosed with not IPA

cThe optimal cutoff value of the BAL GM assay from the ROC curve according to Youden's index.

Impact of mold-active antifungal agents before BAL

The following mold-active antifungal agents were used before BAL: amphotericin B deoxycholate, liposomal amphotericin B, caspofungin, micafungin, itraconazole, voriconazole and posaconazole. The median time of receipt of a mold-active antifungal agent before BAL was 7 days (interquartile range [IQR], 2.5-18 days). The sensitivity of the BAL GM assay was 48% (95% CI, 27-70%), and the median value of BAL GM assay in proven and probable IPA was 0.96 in patients who received a mold-active antifungal agent before BAL, while the sensitivity of the BAL GM assay was 68% (95% CI, 43-87%), and the median value of BAL GM assay was 2.58 in those who did not received a mold-active antifungal agent, as shown in supplemental Table 2 and supplemental Figure 1. The sensitivity of the BAL GM assay in patients who previously received a mold-active antifungal agent before BAL was numerically lower than that in those who did not previously receive mold-active antifungal agents, but this did not reach any statistical significance (p value = 0.66). The correlation between the number of days from the first receipt of mold-active antifungal agent and the BAL GM titer is shown in supplemental Figure 2. The scatterplots had a positive slope, but there was no significant correlation (p value = 0.49).

Table 4. Diagnostic performance of the BAL GM assay by previous receipt of mold-active antifungal agent in patients with suspected IPA who had negative serum GM results

Proven and Probable IPA vs Not IPA	Sensitivity % (n/N [a], 95% CI)	Specificity % (n/N [b],95% CI)	PPV % (95% CI)	NPV % (95% CI)	Positive Likelihood ratio (95% CI)	Negative Likelihood ratio (95% CI)
Mold-active antifungal agent before BAL	48[c] (11/23, 27-69)	91 (9/98, 83-96)	55 (36-72)	88 (83-92)	5.2 (2.5-11)	0.57 (0.39-0.85)
No mold-active antifungal agent before BAL	68[c] (13/19, 43-87)	95 (89/94, 88-98)	72 (51-87)	94 (88-97)	13 (5.2-32)	0.33 (0.17-0.65)

Abbreviation: CI, confidence interval; PPV, positive predictive value; NPV, negative predictive value

aNumber of patients with a positive test result/number of patients tested among those diagnosed with proven and probable IPA

bNumber of patients with a negative test result/number of patients tested among those diagnosed with not IPA

cThere was no significant difference in the sensitivity of the BAL GM assay between those with prior receipt of mold-active antifungal agents before BAL and the comparator (p value = 0.66).

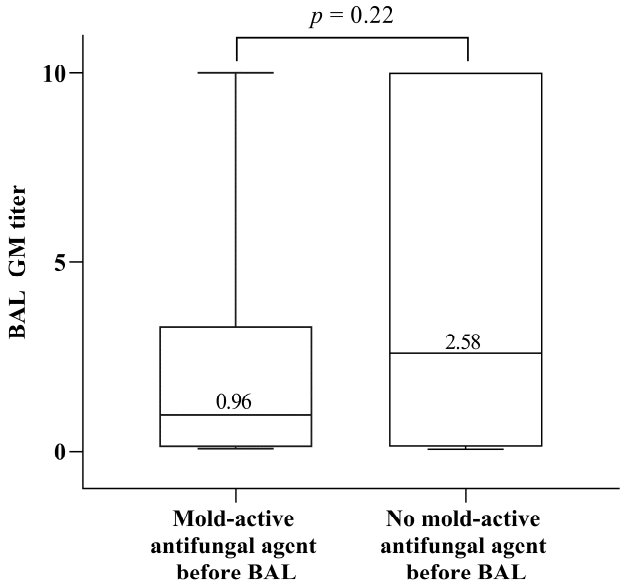


Figure 3. Box-and-whisker plot showing the BAL GM titer by prior receipt of mold-active antifungal agents. The boxes indicate lower and upper quartiles, central lines indicate the median, and the ends of the whiskers indicate the minimum and maximum.

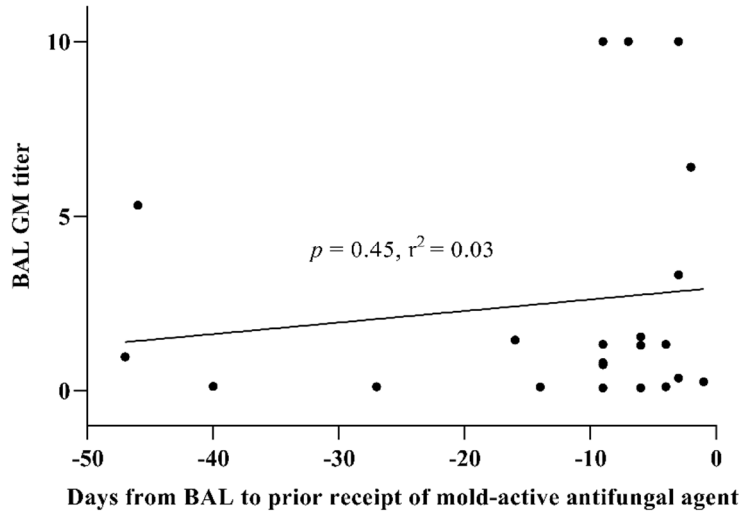


Figure 4. Scatterplot by Spearman's correlation between the days from first receipt of mold-active antifungal agent and the BAL GM titer

Other (opportunistic) infections additionally identified by BAL

Other infections identified by BAL in this study are shown in Table 1. Of 190 patients who underwent diagnostic tests for *P. jirovecii* by BAL, 26 (14%) were identified as PCP. Of 188 patients who had CMV culture results or quantitative PCR titer of CMV DNA from BAL fluid, 9 (5%) were diagnosed as CMV pneumonia. Also, among 193 patients with respiratory virus PCR results from BAL fluid, 12 (6%) showed various respiratory viral infection only from BAL fluid.

DISCUSSION

In this hematology unit-based retrospective study comprising 234 patients with suspected IPA, our results showed that sequential BALs in patients with negative serum GM assay results provided additional diagnostic yield in more than half of patients. In addition, approximately one quarter of patients with suspected IPA had additional evidence of other opportunistic infections from BAL. Therefore, our findings can provide important information for clinician decision making on the rationale of performing a BAL in patients with suspected IPA with negative serum GM assay results.

Regarding the demand for a prompt and sensitive diagnostic tool, a serum GM assay has been widely used as complementary to a sputum fungal culture. However, the sensitivity of the serum GM assay for detecting IPA of approximately 65% [3] is still suboptimal. As a result, the possibility of IPA in patients with negative serum GM assay results should be considered in the setting of classical host factors, and BAL GM is recommended for as an additional invasive test [12]. However, there are limited data about the additional benefit of a BAL GM assay in patients with suspected IPA with negative serum GM results. Our study focused on this clinical question and identified that sequential BAL GM assays performed in patients with negative serum GM results provided additional diagnostic yield in 57%.

It is worth noting that the sensitivity of the BAL GM assay in our study was 57% and is lower than that reported in previous studies. In the study by Maertens et al, the sensitivity of the BAL GM assay in patients with hematologic disease was 91% at a cutoff value of 1.0 [4], and in another prospective study involving ICU patients, the sensitivity of the BAL GM assay was 88% at a cutoff value of 0.5 [13]. A possible explanation for this finding is that prior receipt of a mold-active antifungal agent could have influenced the BAL GM titer. Mold-active antifungal agents are well-known factors affecting both serum GM assays [14-16] and BAL GM assays [17]. In a study involving patients with hematological diseases, 71% of patients with proven and probable IPA received mold-active antifungal agents before BAL, and antifungal therapy administered for ≥2 days significantly decreased the sensitivity of the BAL GM assay from 79% to 50% at a cutoff value of 0.5. [17]. Likewise, approximately half of patients with proven and probable IPA received mold-active antifungal agents before BAL in our study, and the sensitivity of the BAL GM assay decreased from 68% to 48% with the use of a mold-active antifungal agent before BAL (Supplemental Table 2). However, we did not find any statistically significant association of prior antifungal use with the sensitivity of the BAL GM assay ($p = 0.66$, Supplemental Table 2) or with the quantitative level of BAL GM ($p = 0.22$, Supplemental Figure 1). Therefore, there may be more factors affecting the sensitivity of the BAL GM assay in real clinical practice; thus, further studies are needed on this area.

Furthermore, we found that approximately one-quarter of patients with suspected IPA had other

(opportunistic) infections including P. jirovecii, CMV or respiratory viral infections (Table 1). Since IPA, PCP, and CMV pneumonia share similar risk factors such as hematologic malignancy, hematopoietic stem cell transplantation, and high-dose steroid and immunosuppressant use [18-20], differential diagnosis for these infections is critical to start the most appropriate drug. As a result, considering this additional benefit of detecting other opportunistic infections, the recommendation for BAL in patients with suspected IPA who have negative GM results is warranted to detect other opportunistic infections or coinfections.

This study had several limitations. First, only 4 proven cases of IPA were included in our study, and no autopsies were performed. Therefore, some misclassification bias could be possible. In addition, the definition of probable IPA included positive BAL GM results, so some verification bias might have been present. Second, routine PCR for Aspergillus spp. was not possible in out hospital during the study period; therefore, Aspergillus PCR was not considered for the diagnosis of IPA in our study. Thus, some IPA cases may have been missed or misclassified. Third, we excluded patients with possible IPA from the final analysis. Therefore, some repeated GM-negative IPA cases may have been excluded, and the estimation of the BAL GM assay might have been overestimated.

In conclusion, our results suggest that sequential BALs in patients with suspected IPA who had initial negative serum GM results provided additional diagnostic yield in approximately half of patients with evidence of other coinfections.

REFERENCES

1. Kousha, M., R. Tadi, and A.O. Soubani, Pulmonary aspergillosis: a clinical review. European Respiratory Review, 2011. **20**(121): p. 156-174.
2. Tarrand, J.J., et al., Diagnosis of invasive septate mold infections. A correlation of microbiological culture and histologic or cytologic examination. Am J Clin Pathol, 2003. **119**(6): p. 854-8.
3. Pfeiffer, C.D., J.P. Fine, and N. Safdar, Diagnosis of Invasive Aspergillosis Using a Galactomannan Assay: A Meta-Analysis. Clinical Infectious Diseases, 2006. **42**(10): p. 1417-1727.
4. Maertens, J., et al., Bronchoalveolar lavage fluid galactomannan for the diagnosis of invasive pulmonary aspergillosis in patients with hematologic diseases. Clin Infect Dis, 2009. **49**(11): p. 1688-93.
5. Segal, B.H. and T.J. Walsh, Current approaches to diagnosis and treatment of invasive aspergillosis. Am J Respir Crit Care Med, 2006. **173**(7): p. 707-17.
6. Donnelly, J.P., et al., Revision and Update of the Consensus Definitions of Invasive Fungal Disease From the European Organization for Research and Treatment of Cancer and the Mycoses Study Group Education and Research Consortium. Clin Infect Dis, 2019.
7. Fauchier, T., et al., Detection of Pneumocystis jirovecii by Quantitative PCR To Differentiate Colonization and Pneumonia in Immunocompromised HIV-Positive and HIV-Negative Patients. J Clin Microbiol, 2016. **54**(6): p. 1487-1495.
8. Ljungman, P., et al., Definitions of Cytomegalovirus Infection and Disease in Transplant Patients for Use in Clinical Trials. Clinical Infectious Diseases, 2016. **64**(1): p. 87-91.
9. Boeckh, M., et al., Cytomegalovirus (CMV) DNA Quantitation in Bronchoalveolar Lavage Fluid From Hematopoietic Stem Cell Transplant Recipients With CMV Pneumonia. The Journal of infectious diseases, 2017. **215**(10): p. 1514-1522.
10. Kim, M.C., et al., CT findings in viral lower respiratory tract infections caused by parainfluenza virus, influenza virus and respiratory syncytial virus. Medicine (Baltimore), 2016. **95**(26): p. e4003.
11. Ruopp, M.D., et al., Youden Index and optimal cut-point estimated from observations affected by a lower limit of detection. Biometrical journal. Biometrische Zeitschrift, 2008. **50**(3): p. 419-430.
12. Patterson, T.F., et al., Practice Guidelines for the Diagnosis and Management of Aspergillosis: 2016 Update by the Infectious Diseases Society of America. Clinical Infectious Diseases, 2016. **63**(4): p. e1-e60.

13. Meersseman, W., et al., Galactomannan in bronchoalveolar lavage fluid: a tool for diagnosing aspergillosis in intensive care unit patients. Am J Respir Crit Care Med, 2008. **177**(1): p. 27-34.
14. Jung, J., et al., Clinical characteristics, radiologic findings, risk factors and outcomes of serum galactomannan-negative invasive pulmonary aspergillosis. J Microbiol Immunol Infect, 2018. **51**(6): p. 802-809.
15. Marr, K.A., et al., Detection of Galactomannan Antigenemia by Enzyme Immunoassay for the Diagnosis of Invasive Aspergillosis: Variables That Affect Performance. The Journal of Infectious Diseases, 2004. **190**(3): p. 641-649.
16. Marr, K.A., et al., Antifungal therapy decreases sensitivity of the Aspergillus galactomannan enzyme immunoassay. Clin Infect Dis, 2005. **40**(12): p. 1762-9.
17. Racil, Z., et al., Galactomannan detection in bronchoalveolar lavage fluid for the diagnosis of invasive aspergillosis in patients with hematological diseases-the role of factors affecting assay performance. Int J Infect Dis, 2011. **15**(12): p. e874-81.
18. Konoplev, S., et al., Cytomegalovirus pneumonia in adult autologous blood and marrow transplant recipients. Bone Marrow Transplant, 2001. **27**(8): p. 877-81.
19. Yale, S.H. and A.H. Limper, Pneumocystis carinii pneumonia in patients without acquired immunodeficiency syndrome: associated illness and prior corticosteroid therapy. Mayo Clin Proc, 1996. **71**(1): p. 5-13.
20. Sepkowitz, K.A., et al., Pneumocystis carinii pneumonia among patients without AIDS at a cancer hospital. Jama, 1992. **267**(6): p. 832-7.

국문요약

혈청 갈락토만난이 음성으로 확인된 침습성 폐 아스페르길루스증 의심환자에서 기관지 폐포 세척액 갈락토만난의 진단적 유용성

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연구배경: 혈청 갈락토만난이 음성으로 확인된 침습성 폐 아스페르길루스증 의심환자에서 기관지 폐포 세척액 갈락토만난의 진단적 유용성에 대한 데이터는 실제 임상현장에서 부족하다. 따라서 본 연구에서는 침습성 폐 아스페르길루스증이 의심되나 혈청 갈락토만난이 음성인 경우, 기관지 폐포 세척액의 진단적 유용성에 대해 알아보고자 하였다.

방법: 본 연구는 2008 년 5 월부터 2019 년 4 월까지 3차 대학병원에서의 의무기록을 후향적으로 분석하여 이루어졌다. 침습성 폐 아스페르길루스증이 의심되나 혈청 갈락토만난 검사 결과가 음성이고 이후 기관지 폐포 세척술을 받은 환자 모두가 연구에 등록되었다. 환자들은 개정된 2019년 EORTC/MSG 정의에 따라 proven, probable, possible, 그리고 not IPA로 분류되었다.

결과: 4명의 proven IPA, 38명의 probable IPA, 107명의 possible IPA, 192 명의 not IPA를 포함하여 총 341 명의 환자가 연구에 등록되었다. 107명의 possible IPA는 최종 분석에서 제외하였다. 혈청 갈락토만난 결과가 음성이었던 proven 또는 probable IPA 환자 42 명 중 24 명 (57 %)은 기관지 폐포 세척액 갈락토만난 결과가 양성으로 확인되었고, 기관지 폐포 세척액 갈락토만난 결과가 음성으로 확인된 나머지 18명 (43 %) 중 2명 (5 %)은 조직학적 검사로 proven IPA, 6명 (14 %)은 반복적인 객담 곰팡이 배양으로 probable IPA, 그리고 12명 (29%)은 기관지 폐포 세척술 시행 이후 반복적인 혈청 갈락토만난 검사로 probable IPA로 진단되었다. 192명의 not IPA 환자 중에서 14명 (7%)은 기관지 폐포 세척액 갈락토만난 양성 (n = 14) 또는 기관지 폐포 세척액 곰팡이 배양에서 양성 (n = 8)으로 나타났다. 추가적으로 기관지 폐포 세척술을

통해 폐포자충 폐렴 (14 % [26/190]), 거대세포바이러스 폐렴 (5 % [9/188]), 호흡기 바이러스성 폐렴 (6 % [12/193])을 포함한 다른 공동 감염을 확인할 수 있었다.

결론: 초기 혈청 갈락토만난 검사에서 음성으로 확인되나 침습성 폐 아스페르길루스증이 의심되는 환자에서, 기관지 폐포 세척액에서 시행한 갈락토만난은 절반 이상의 환자에서 추가적인 진단적 이득이 있었을 뿐 아니라, 다른 공동 감염의 증거 역시 제공하였다.

중심단어: 침습성 폐 아스페르길루스증, 기관지 폐포 세척술, 갈락토만난