Performance of Galactomannan, Beta-d-Glucan, Aspergillus Lateral-Flow Device, Conventional Culture, and PCR Tests with Bronchoalveolar Lavage Fluid for Diagnosis of Invasive Pulmonary Aspergillosis


Updated information and services can be found at: http://jcm.asm.org/content/52/6/2039

**REFERENCES**

This article cites 53 articles, 29 of which can be accessed free at: http://jcm.asm.org/content/52/6/2039#ref-list-1

**CONTENT ALERTS**

Receive: RSS Feeds, eTOCs, free email alerts (when new articles cite this article), more»
Invasive fungal infections (IFI) remain an important cause of morbidity and mortality among immunocompromised patients. Invasive pulmonary aspergillosis (IPA) is caused by Aspergillus fumigatus and other Aspergillus species. Being an abundant component of inhaled air, these organisms represent one of the leading causes of IFI-related morbidity and mortality (1–5). Due to the crude mortality of 80 to 90% in the absence of adequate treatment, timely diagnosis and early start of antifungal therapy are key factors in the successful treatment of IPA, as delayed antifungal therapy has a negative impact on the survival of these patients (6). Various studies have shown that early diagnosis and initiation of antifungal therapy may improve IFI survival to >80% (7–9). Diagnosing IPA, however, remains difficult, as clinical signs and symptoms, as well as radiological findings, are often unspecific, and conventional culture methods display low sensitivities (8). In a large autopsy-based retrospective analysis, only 25% of patients with IFI as confirmed by autopsy were identified premortem by culture-based methods, underlining the difficulty in diagnosing IPA by conventional methods and emphasizing the need for novel serological and molecular markers (10, 11). In recent years, antigen testing has therefore become one of the cornerstones of IFI diagnostics (12, 13). Galactomannan (GM) is a polysaccharide component of the cell wall of Aspergillus spp. that is released into the bloodstream by growing hyphae and germinating spores/conidia. The diagnostic performance of GM testing with bronchoalveolar lavage (BAL) fluid specimens is promising, as it has higher sensitivity than that of serum testing (14). The test, however, has several limitations. As false-positive results may occur, patient factors, such as comorbidities (e.g., β-lactam antibacterials), underlying diseases, host factors (e.g., renal failure), diagnostic imaging, clinical signs, and former medication(s) must be taken into account to correctly interpret GM levels (15–17). Furthermore, some studies have shown that the sensitivity of the GM test decreases significantly in the case of the administration of antifungal prophylaxis/empirical therapy, whereas other reports have shown its usefulness for diagnosing breakthrough IFI (13, 18–20).

Another target antigen for IFI diagnosis is β-D-glucan (BDG), a cell wall component of most pathogenic fungi (e.g., Aspergillus

Galactomannan detection in bronchoalveolar lavage (BAL) fluid samples (GM test) is currently considered the gold standard test for diagnosing invasive pulmonary aspergillosis (IPA). The limitations, however, are the various turnaround times and availability of testing. We compared the performance of GM testing with that of conventional culture, an Aspergillus lateral-flow-device (LFD) test, a beta-D-glucan (BDG) test, and an Aspergillus PCR assay by using BAL fluid samples from immunocompromised patients. A total of 78 BAL fluid samples from 78 patients at risk for IPA (74 samples from Graz and 4 from Mannheim) collected between December 2012 and May 2013 at two university hospitals in Austria and Germany were included. Three patients had proven IPA, 14 probable IPA, and 17 possible IPA, and 44 patients had no IPA. The diagnostic accuracies of the different methods for probable/proven IPA were evaluated. The diagnostic odds ratios were the highest for the GM, PCR, and LFD tests. The sensitivities for the four methods (except culture) were between 70 and 88%. The combination of the GM (cutoff optical density index [ODI], >1.0) and LFD tests increased the sensitivity to 94%, while the combination of the GM test (>1.0) and PCR resulted in 100% sensitivity (specificity for probable/proven IPA, 95 to 98%). The performance of conventional culture was limited by low sensitivity, while that of the BDG test was limited by low specificity. We evaluated established and novel diagnostic methods for IPA and found that the Aspergillus PCR, LFD, and GM tests were the most useful methods for diagnosing the disease by using BAL fluid samples. In particular, the combination of the GM test and PCR or, if PCR is not available, the LFD test, allows for sensitive and specific diagnosis of IPA.

Performance of Galactomannan, Beta-D-Glucan, Aspergillus Lateral-Flow Device, Conventional Culture, and PCR Tests with Bronchoalveolar Lavage Fluid for Diagnosis of Invasive Pulmonary Aspergillosis

M. Hoenigl,⁎ a J. Prattes, a B. Spiess, b J. Wagner, a F. Prueller, a R. B. Raggam, a V. Posch, a W. Duettmann, d K. Hoenigl, a A. Wölfle, d C. Koidl, e W. Buzina, e M. Reinwald, b C. R. Thornton, f R. Krause, a D. Buchheidt e

Section of Infectious Diseases and Tropical Medicine, Medical University of Graz, Graz, Austria; Department of Hematology and Oncology, Mannheim University Hospital, University of Heidelberg, Mannheim, Germany; Clinical Institute of Medical and Chemical Laboratory Diagnostics, Medical University of Graz, Graz, Austria; Division of Hematology, Medical University of Graz, Graz, Austria; Institute of Hygiene, Microbiology and Environmental Medicine, Medical University of Graz, Graz, Austria; Biosciences, University of Exeter, Exeter, United Kingdom; Division of Pulmonology, Medical University of Graz, Graz, Austria

doi:10.1128/JCM.00467-14

Copyright © 2014, American Society for Microbiology. All Rights Reserved.
Based on the detection of Aspergillus antigens by the monoclonal antibody (MAb) IF5, can be performed easily in every laboratory using BAL fluid or serum specimens, and it has a time to results of approximately 15 min. Recent studies have shown the immense potential of this test with human BAL fluid and serum samples (30–32) but have been limited by the small sample sizes, especially for BAL fluid samples (30, 33). More extensive clinical studies are therefore needed to evaluate this new point-of-care device in different patient cohorts.

Even though sensitivity may decrease while on antifungal prophylaxis, PCR has been shown to be a very promising method for diagnosing fungal infection by using BAL fluid specimens from immunocompromised patients (34–37), especially when combined with the GM test (38). Despite major concerns, such as the lack of external standardization, the variety of different methods employed, and the recognized need for larger studies, PCR diagnostics using BAL fluid specimens seems to be a promising approach for the diagnosis of IFI. Harmonization efforts are ongoing to enable the inclusion of PCR in the next revision of the EORTC classifications (35, 39).

In this study, we compared the diagnostic performances for IPA of the GM test, conventional culture, Aspergillus LFD test, BDG test, and Aspergillus PCR assay as single and combined diagnostic tools when used with BAL fluid samples from immunocompromised patients.

**MATERIALS AND METHODS**

This prospective, part retrospective study used 78 routine BAL fluid samples from 78 adult immunocompromised patients (74 samples from Graz and 4 from Mannheim) that were tested in a routine clinical setting for GM as well as with conventional mycological culture between July 2012 and May 2013 at the two university hospitals of Graz, Austria, and Mannheim, Germany. IPA was graded in accordance with the revised criteria by the European Organization for Research and Treatment of Cancer Invasive Fungal Infections Cooperative Group (EORTC) and the Mycoses Study Group (MSG) of the National Institute of Allergy and Infectious Disease (38, 40).

In 67 samples (63 from Graz and 4 from Mannheim), LFD, BDG, and PCR testing were performed in addition to GM testing and mycological culture. In 11 additional samples (all from Graz), there was insufficient BAL fluid material for PCR testing; therefore, only LFD and BDG testing were performed in addition to GM testing and mycological culture. In contrast to BAL fluid, serum GM and BDG levels were available from approximately two-thirds of patients only, and so these results were excluded from this study.

Conventional mycological culture was routinely and prospectively performed in Graz (microbiology laboratory, Department of Internal Medicine) and Mannheim (Institute of Medical Microbiology and Hygiene, Mannheim University Hospital). BAL fluid GM concentrations were prospectively determined in a routine clinical setting by the Platelia enzyme immunoassay (EIA) (Bio-Rad Laboratories) in Graz (Institute of Hygiene, Microbiology, and Environmental Medicine) and Mannheim (Institute of Medical Microbiology and Hygiene, Mannheim University Hospital). For GM testing, we used optical density index (ODI) cutoffs of 0.5 and 1.0 to evaluate the diagnostic performance of the test.

LFD testing was prospectively performed at the microbiology laboratory, Department of Internal Medicine, Medical University of Graz, and the scientific laboratory, Department of Hematology and Oncology, Mannheim University Hospital. The LFD test is based on the detection of an Aspergillus antigen by MAb IF5. The target antigen is an extracellular glycoprotein that is exclusively secreted during active growth of the fungus and represents a surrogate marker of Aspergillus infection (30). The monoclonal antibody IF5 has been incorporated into an immunochromatographic assay (a point-of-care diagnostic tool), which is easy to use. The rapidity of the test using BAL fluid samples (the time to results is only 15 min) is particularly remarkable. The IF5 LFD results in qualitative data based on the test line intensity, ranging from strong positive (++) to weak positive (+) to negative (−). As the test is read by the naked eye, the test interpretation depends on a subjective evaluation. Wiederhold and colleagues (41) have shown, however, that the results are reproducible between different laboratories and studies. Regardless of the test line intensity, however, all positive test results with BAL fluid samples indicate the germination of spores and the development of potentially pathogenic hyphae in the lungs (30). Testing was performed according to the manufacturer’s instructions. For BAL fluid testing, 100 μl of BAL fluid sample was applied to the LFD, without having received pretreatment (30). The results were read after 15 min and interpreted in line with those described in previous publications (30).

After performing culture, GM, and LFD testing, all 78 samples were frozen and stored at −70°C for retrospective BDG and PCR testing. BDG testing was performed retrospectively in all 78 samples at the Clinical Institute of Medical and Chemical Laboratory Diagnostics, Medical University of Graz, using the commercial available Fungitell assay (Cape Cod Diagnostics) with an adopted protocol suitable for use on a routine BCS XP coagulation analyzer, as described previously (42). For BDG testing, we used the cutoffs of 80 pg/ml and 200 pg/ml to evaluate the diagnostic performance of the test.

A nested Aspergillus PCR assay was performed retrospectively in 67 samples at the scientific lab of the University Hospital of Mannheim, according to the protocol of Skladny et al. (43), as described previously.

Our study was conducted in accordance with the Declaration of Helsinki, 1996, good clinical practices, and applicable local regulatory requirements and laws. The study protocol was approved by the local ethics committee, Medical University Graz, Austria (EC-no. 25-221 ex 12/13) and registered at ClinicalTrials.gov (identifier NCT02058316).

Statistical analysis was performed using SPSS, version 20 (SPSS, Inc., Chicago, IL, USA). Negative predictive value (NPV), positive predictive value (PPV), sensitivity, and specificity were calculated where applicable. The different diagnostic methods were compared using the diagnostic odds ratio (DOR) method. All DOR values were displayed with 95% confidence intervals (95% CI). The specificity, PPV, NPV, and DOR were calculated three times (i) for probable/proven IPA versus no IPA (exclusion of possible IPA cases), (ii) for probable/proven IPA versus possible/no IPA, and (iii) for possible/probable IPA versus no IPA. This was necessary, as cases with possible IPA might in fact have had IPA or not, while IPA can almost always be excluded in patients that do not fulfill the criteria at all. Receiver operating characteristic (ROC) curve analysis was performed for BDG and GM levels. Area under the curve (AUC) values were displayed, including 95% confidence intervals (CI).

**RESULTS**

A total of 78 patients (54 males, 24 females; median age, 58 years; range, 24 to 77 years) were included in the study. The majority...
TABLE 1 Demographic data and underlying diseases of patients with GM test, LFD test, BDG test, and mycological culture results

<table>
<thead>
<tr>
<th>Patient characteristics</th>
<th>With PCR results</th>
<th>Without PCR results</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>67</td>
<td>11</td>
</tr>
<tr>
<td>No. (%) of males/no. (%) of females</td>
<td>45 (67)/22 (33)</td>
<td>9 (82)/2 (18)</td>
</tr>
<tr>
<td>Age median (range) (yr)</td>
<td>58 (24–77)</td>
<td>55 (34–67)</td>
</tr>
</tbody>
</table>

Primary underlying disease/condition (no. [%])

- Hematological malignancy: 43 (64), 4 (36)
- AML: 23 (34), 2 (18)
- NHL: 7 (10), 2 (18)
- Solid organ transplantation: 4 (6), 7 (64)
- Underlying chronic lung disease: 12 (18)
- ICU (sepsis): 4 (6)
- AIDS: 2 (3)
- Chronic autoimmune disease: 2 (3)

a GM, galactomannan; BDG, beta-d-glucan.

(47/78 [60%]) of patients had underlying hematological malignancies, 11/78 had undergone solid organ transplantation, 12/78 had underlying chronic lung diseases (including high-grade chronic obstructive pulmonary disease [COPD] and pulmonary sarcoidosis), 4/78 had sepsis treated in intensive care unit (ICU), 2/78 patients had AIDS, and 2/78 had chronic autoimmune diseases (under immunosuppressive therapies). The demographic data and underlying diseases of patients with/without PCR results are depicted in Table 1.

In 67 samples, all five methods (including PCR) were compared. Out of these 67 patients, three had proven IPA, seven probable IPA, 16 possible IPA, and 41 no IPA. The sensitivities, specificities, PPVs, NPVs, and DORs of PCR (alone or in combination) for probable/proven IPA versus no IPA were as follows: PCR, 96%, 78%, 95%, and 60.7 (95% CI, 8.6 to 429); PCR and/or a BDG level of >80 pg/ml, 63%, 30%, 97%, and 15.4 (95% CI, 1.8 to 130); PCR and/or LFD test, 81%, 45%, 98%, and 37.7 (95% CI, 4.3 to 329); PCR and/or a GM ODI of >1.0, 95%, 77%, 100%, and 327 (95% CI, 15.7 to 6,809). The respective sensitivities, PPVs, NPVs, and DORs of PCR (alone or in combination) for possible/probable IPA versus no IPA were as follows: PCR, 35%, 100%, 71%, and 45 (95% CI, 2.5 to 817); PCR and/or a BDG level of >80 pg/ml, 77%, 67%, 84%, and 10.3 (95% CI, 3.2 to 33); PCR and/or the LFD test, 69%, 90%, 83%, and 44 (95% CI, 8.5 to 228); and PCR and/or a GM ODI of >1.0, 46%, 92%, 74%, and 34.3 (95% CI, 4.1 to 288).

In another 11 patients, four methods (except PCR) were compared. Seven of these 11 patients had probable IPA, one possible, and three no IPA. The sensitivities, specificities, PPVs, NPVs, and DORs of the GM, BDG, and LFD tests, mycological culture, and three combinations (GM ODI of >1.0 and/or positive LFD test, BDG level of >80 pg/ml and/or GM ODI of >1.0, BDG level of >80 pg/ml and/or positive LFD test) for probable/proven IPA in all 78 patients (including the abovementioned 11) are depicted in Table 3. The respective specificities, PPVs, NPVs, and DORs for probable/proven IPA versus possible/no IPA were as follows: GM ODI of >1.0, 95%, 77%, 95%, and 74 (95% CI, 15.4 to 429); BDG level of >80 pg/ml, 72%, 41%, 90%, and 28.9 (95% CI, 7.2 to 133); mycological culture, 97%, 82%, 88%, and 33.2 (95% CI, 6.1 to 182); a BDG level of >80 pg/ml, 41%, 95%, and 13.3 (95% CI, 2.8 to 64); a BDG level of >200 pg/ml, 72%, 41%, 90%, and 6.2 (95% CI, 1.9 to 20); LFD test, 84%, 60%, 96%, and 38.3 (95% CI, 2.8 to 194); a BDG level of >80 pg/ml and/or GM ODI of >1.0, 64%, 42%, 98%, and 28.4 (95% CI, 3.5 to 229); a BDG level of >80 pg/ml and/or the LFD test, 56%, 38%, 94%, and 9.4 (95% CI, 2 to 44.9); and a GM ODI of >1.0 and/or the LFD test, 82%, 59%, 98%, and 73 (95% CI, 8.7 to 608). The respective sensitivities, PPVs, NPVs, and DORs for possible/probable IPA versus Table 2 Diagnostic performance of BAL fluid GM test, culture, BDG test, LFD test, and PCR for probable and proven IPA versus no IPA in patients with all test results available

<table>
<thead>
<tr>
<th>Test method and/or conditions</th>
<th>Sensitivity (n = 10)</th>
<th>Specificity (n = 41)</th>
<th>PPV</th>
<th>NPV</th>
<th>DOR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GM ODI of &gt;1.0</td>
<td>70</td>
<td>98</td>
<td>88</td>
<td>93</td>
<td>93.3 (8.5-1,030)</td>
</tr>
<tr>
<td>GM ODI of &gt;0.5</td>
<td>80</td>
<td>98</td>
<td>89</td>
<td>95</td>
<td>160 (12.9-1,984)</td>
</tr>
<tr>
<td>Mycological culture</td>
<td>50</td>
<td>95</td>
<td>71</td>
<td>89</td>
<td>19.5 (3-129)</td>
</tr>
<tr>
<td>BDG &gt; 80 pg/ml</td>
<td>80</td>
<td>76</td>
<td>44</td>
<td>94</td>
<td>12.4 (2.3-68)</td>
</tr>
<tr>
<td>BDG &gt; 200 pg/ml</td>
<td>60</td>
<td>88</td>
<td>55</td>
<td>90</td>
<td>10.8 (2.5-52)</td>
</tr>
<tr>
<td>LFD test</td>
<td>80</td>
<td>95</td>
<td>80</td>
<td>95</td>
<td>78 (9.5-639)</td>
</tr>
<tr>
<td>PCR</td>
<td>70</td>
<td>100</td>
<td>100</td>
<td>93</td>
<td>161 (7.5-3,445)</td>
</tr>
<tr>
<td>PCR and/or BDG &gt; 80 pg/ml</td>
<td>90</td>
<td>76</td>
<td>47</td>
<td>97</td>
<td>27.9 (3.1-248)</td>
</tr>
<tr>
<td>PCR and/or LFD test</td>
<td>90</td>
<td>95</td>
<td>82</td>
<td>98</td>
<td>176 (14.3-2154)</td>
</tr>
<tr>
<td>GM ODI of &gt;1.0 and/or PCR</td>
<td>100</td>
<td>98</td>
<td>91</td>
<td>99</td>
<td>567 (21.5-14,946)</td>
</tr>
<tr>
<td>GM ODI of &gt;1.0 and/or LFD test</td>
<td>90</td>
<td>93</td>
<td>75</td>
<td>97</td>
<td>114 (10.6-1,228)</td>
</tr>
</tbody>
</table>

a GM, galactomannan; BDG, beta-d-glucan; LFD, lateral-flow-device.
b OD, optical density index.
c PPV, positive predictive value; NPV, negative predictive value.
d DOR, diagnostic odds ratio; 95% CI, 95% confidence interval.

TABLE 2 Diagnostic performance of BAL fluid GM test, culture, BDG test, LFD test, and PCR for probable and proven IPA versus no IPA in patients with all test results available
TABLE 3 Diagnostic performance of BAL fluid GM test, culture, BDG test, and LFD test for probable and proven IPA versus no IPA in all patients included

<table>
<thead>
<tr>
<th>Test method and/or conditions</th>
<th>Performance characteristic (%)</th>
<th>DOR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sensitivity (n = 17)</td>
<td>Specificity (n = 44)</td>
</tr>
<tr>
<td>GM ODI of &gt;3.0</td>
<td>41</td>
<td>100</td>
</tr>
<tr>
<td>GM ODI of &gt;1.0</td>
<td>71</td>
<td>98</td>
</tr>
<tr>
<td>GM ODI of &gt;0.5</td>
<td>82</td>
<td>95</td>
</tr>
<tr>
<td>Mycological culture</td>
<td>53</td>
<td>95</td>
</tr>
<tr>
<td>BDG &gt; 80 pg/ml</td>
<td>88</td>
<td>73</td>
</tr>
<tr>
<td>BDG &gt; 200 pg/ml</td>
<td>71</td>
<td>84</td>
</tr>
<tr>
<td>LFD test</td>
<td>88</td>
<td>95</td>
</tr>
<tr>
<td>BDG &gt; 80 pg/ml and/or GM ODI of &gt;1.0</td>
<td>94</td>
<td>73</td>
</tr>
<tr>
<td>BDG &gt; 80 pg/ml and/or LFD test</td>
<td>88</td>
<td>68</td>
</tr>
<tr>
<td>GM ODI of &gt;1.0 and/or LFD test</td>
<td>94</td>
<td>93</td>
</tr>
</tbody>
</table>

a GM, galactomannan; BDG, beta-D-glucan; LFD, lateral-flow-device.

b ODI, optical density index.

c PPV, positive predictive value; NPV, negative predictive value.

DOR, diagnostic odds ratio; 95% CI, 95% confidence interval.

Discussion

We evaluated established and novel diagnostic methods for IPA and found that GM, Aspergillus PCR, and the LFD tests had the highest diagnostic potential with BAL fluid samples. With respect to combinations, GM testing (with a cutoff ODI of 1.0) and/or positive PCR, or a combination of the GM test (>1.0 ODI) and/or a positive LFD test exhibited high sensitivities (>90%) as well as the highest DORs. The performance of conventional culture was limited by its low sensitivity, while that of the BDG test was limited by low specificity.

The BAL fluid GM test is the current gold standard for diagnosing IPA, as it was reported to have an impact on patient survival (2, 44, 45). However, recommendations for optimal GM cutoff ODIs from BAL fluid specimens vary widely, between 0.5 and 3.0 (44-48). In this study, not surprisingly, sensitivity decreased with a higher cutoff value (from 82% at 0.5 ODI to 71% at 1.0 ODI to 41% at 3.0 ODI) and specificity increased. Overall, DOR showed that 1.0 ODI might be as useful as a cutoff as 0.5 ODI; also, the AUCs in the ROC curve analysis were nearly identical for both cutoff values.

In our cohort, Aspergillus PCR from BAL fluid was a promising single test method with a sensitivity of 70%, a near-to-perfect specificity, and convincing DORs. While in this study, a well-evaluated PCR protocol was used (35, 36, 46, 49, 50), it has to be emphasized that the results of our study are not transferable to other PCR assays, as the performances differ of the multiple PCR methods currently in use, and they also lack external standardization. Another drawback of PCR testing in general is the amount of specimen needed to perform valid testing (about 2 ml), which is markedly more material than that needed for the GM (600 μl, including serial retesting), BDG (about 200 μl, including serial retesting), and the LFD (100 μl) tests. For this reason, we were not able to perform PCR for all of the samples.
Another promising method is the *Aspergillus* LFD test, with a sensitivity of 88% and specificity between 84% and 95%. Recent single-center studies, including one from our center, have shown the potential of this test for diagnosing IPA using human BAL fluid and serum samples (30–33, 37, 51). The high NPV (95% to 96% in this study) may be particularly valuable. The test may therefore be a valuable tool for enabling immediate treatment decisions, as the time to results is only 15 min. Due to the high NPV, LFD testing may not only facilitate early diagnosis but also may prevent overtreatment, which has become frequent (52).

In a recent study, the combination of GM and PCR testing from blood samples has been reported to be effective in directing treatment and in reducing the use of empirical antifungal treatment (53). With respect to combination tests with BAL fluid samples, GM testing with a cutoff 1.0 OD1 and/or positive PCR (sensitivity 100%) exhibited the highest DOR, followed by the combination of a GM OD1 of >1.0 and/or a positive LFD test result (sensitivity, 94%).

Mycological culture of BAL fluid is indispensable, as antifungal susceptibility testing is currently available from culture only. Culture may also grow other molds, like *Mucorales*, which do not result in positive GM or BDG levels or LFD test results. It has to be emphasized, however, that the low sensitivity of conventional culture is its major drawback. Lass-Flörl and colleagues (8) have reemphasized, however, that the low sensitivity of conventional culture may also grow other molds, like *Aspergillus*. Susceptibility testing is currently available from culture only. Culture from blood samples has been reported to be effective in directing treatment and in reducing the use of empirical antifungal treatment and in preventing overtreatment, which has become frequent (52).

In contrast to culture, the BAL fluid BDG test sensitivity was high (for the 80 pg/ml cutoff), but specificity was the major limitation of this test. In our collective, specificity varied between 64% and 84% depending on the cutoff level employed. A meta-analysis conducted by Karageorgopoulos et al. (54) showed an overall sensitivity of the serum BDG assays for IFI diagnosis of approximately 77% (95% confidence interval [CI], 67% to 84%) and a specificity of about 85% (95% CI, 80% to 90%). The PPVs ranged from 59% to 96% and the NPVs reached 95% in some studies. The specificity in BAL fluid samples seems therefore to be lower than that of serum samples, which might be also a result of frequent *Candida* colonization of the upper and lower respiratory tract in critically ill patients. Nevertheless, we found an NPV between 94% and 95% for the 80-pg/ml cutoff, even though our collective did not include *Pneumocystis* infections, for which NPVs of ≥99% have been reported. A negative BDG test result with BAL fluid might therefore still have clinical value.

In conclusion, the GM, *Aspergillus* PCR, and the LFD tests had the highest diagnostic potential for IPA in BAL fluid samples. With regard to combinations, the GM test and PCR or the GM and LFD tests seem to be the most promising. Studies with larger sample sizes are needed to further evaluate these diagnostic approaches.

ACKNOWLEDGMENTS

We acknowledge the help of Brigitte Luttenberger, Brigitta Waitzl, Silke Will, and Natalia Merker in sample organization and preparation, as well as in the performance of the GM test (B. Luttenberger, B. Waitzl) and PCR (S. Will, N. Merker).

The study was funded by an investigator-initiated research grant from Pfizer (W1174981) and supported by funds of the Österreichische Nationalbank (Anniversary Fund, project number 15346). The LFD tests used in this study were provided by C. Thornton, University of Exeter. No other funding was obtained for this study.

M. Hoenigl received a research grant from Merck and Pfizer, served on the speakers’ bureau of Pfizer, Gilead, Astellas, and Merck, and received travel grants from Astellas, Merck, Gilead, and Pfizer. D. Buchheidt is a consultant to Gilead Sciences, received research grants from Gilead Sciences and Pfizer, served on the speakers’ bureau of Astellas, Gilead Sciences, Merck Sharp & Dohme/Merck, and Pfizer, and received travel grants from Astellas, Merck Sharp & Dohme/Merck, and Pfizer. W. Buzina served on the speaker’s bureau of Merck and received travel grants from Pfizer. M. Reinwald received travel grants from Astellas. All other authors have no conflicts of interest.

REFERENCES


Diagnosis of IPA in BAL Fluid


