Performance of the bronchoalveolar lavage fluid later-flow device test in the rapid diagnosis of non-neutropenic invasive pulmonary aspergillosis

Lu-Lu Liu[1], Yu Wang[1], Yu Gu[2], Shan-Shan Li[1], Bi-Lin Chen[1], Kun-Lu Shen[3], Xin Su[1,2,3]

[1]Department of Respiratory and Critical Care Medicine, Jinling Hospital, Medical School of Nanjing University, Nanjing, Jiangsu 210002, China;
[2]Department of Respiratory and Critical Care Medicine, Jinling Hospital, Nanjing Medical University, Nanjing, Jiangsu 210002, China;
[3]Department of Respiratory and Critical Care Medicine, Jinling Hospital, Southern Medical University, Guangzhou, Guangdong 510515, China.

To the Editor: A multicenter retrospective survey in China shows that the most common pulmonary fungal disease is pulmonary aspergillosis.[1] The diagnosis of invasive pulmonary aspergillosis (IPA) remains challenging because of the high false-negative and false-positive rate of galactomannan (GM) test. Domestic research reports that the IPA misdiagnosis rate can be as high as 73%. Besides, GM is time consuming, making the diagnosis delayed.

Recently, a novel lateral-flow device (LFD) had been developed to detect the *Aspergillus* antigen. Furthermore, the *Asp*LFD is a simple operation and point-of-care test. It only needs 15 to 20 min to complete the test. The *Asp*LFD had higher sensitivity in bronchoalveolar lavage fluid (BALF) specimens than serum, the sensitivity of the *Asp*LFD in BALF was 77% to 100%, and the specificity was 81% to 92% in neutropenic IPA patients,[2] which is similar to the BALF GM test.

However, the *Asp*LFD research is still lacking in non-neutropenic IPA patients. Therefore, we performed this study to explore the diagnostic value of *Asp*LFD in non-neutropenic IPA.

The study was approved by the Institute Ethics Committee of Jinling Hospital (No. 2015NJKY-035-04). All participants signed the informed consent. This prospective study was conducted at the Department of Respiratory and Critical Care Medicine, Nanjing Jinling Hospital. Suspected non-neutropenic IPA patients were screened and recruited. A total of 136 BALF samples from 136 patients (76 men, 60 women; median age, 54.9 years; range, 18.0–80.0 years) were collected. Forty-one samples (proven 8; probable 33) fell into IPA group, and ninety-five samples were non-IPA group (bacterial infection, tuberculosis, cancer). Among the samples, thirty-three samples in IPA group and forty-seven samples in non-IPA group were tested for GM [Figure 1A]. The criteria for IPA were based on the European Organization for Research and Treatment of Cancer/Invasive Fungal Infections Cooperative Group and the National Institute of Allergy and Infectious Diseases Mycoses Study Group Consensus Group.[3]

Bronchoscopy was performed according to the standard procedure of Chinese Consensus of BALF in Pulmonary Infectious Diseases. The target bronchus was positioned with computed tomography scans, and 30 to 50 mL saline was injected through the bronchoscope and usually repeated three times. The recovered BALF samples were centrifuged at 3000 × *g* for 10 min to remove blood and mucus, BALF GM enzyme-linked immunosorbent assay (Bio-Rad Laboratories, CA, USA) was performed immediately, and threshold set at 0.7 according to our previous study.[4] The rest BALF was stored in a sterile tube at –80°C for *Asp*LFD (OLM Diagnostics, Newcastle upon Tyne, UK) [Figure 1B]. The BALF samples were vortexed and then centrifuged at 13,000 × g for 1 min, 70 μL of BALF sample was applied to the *Asp*LFD. The results were read after 15 to 30 min. The investigator was blinded to the clinical diagnosis. OLM Diagnostics did not participate in this research. SPSS 22.0 (SPSS Inc., Chicago, IL, USA) was used for statistical analysis.

In our study, more previous tuberculosis and chronic obstructive pulmonary disease in IPA group, and the differences were significant ($P < 0.0001$; $P < 0.05$). Other characteristics between the two groups had no significant differences [Supplementary Table 1, http://links.lww.com/CM9/A215].

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Correspondence to: Xin Su, Department of Respiratory and Critical Care Medicine, Jinling Hospital, Nanjing University, School of Medicine, Nanjing, Jiangsu 210002, China
E-Mail: suxinjs@163.com

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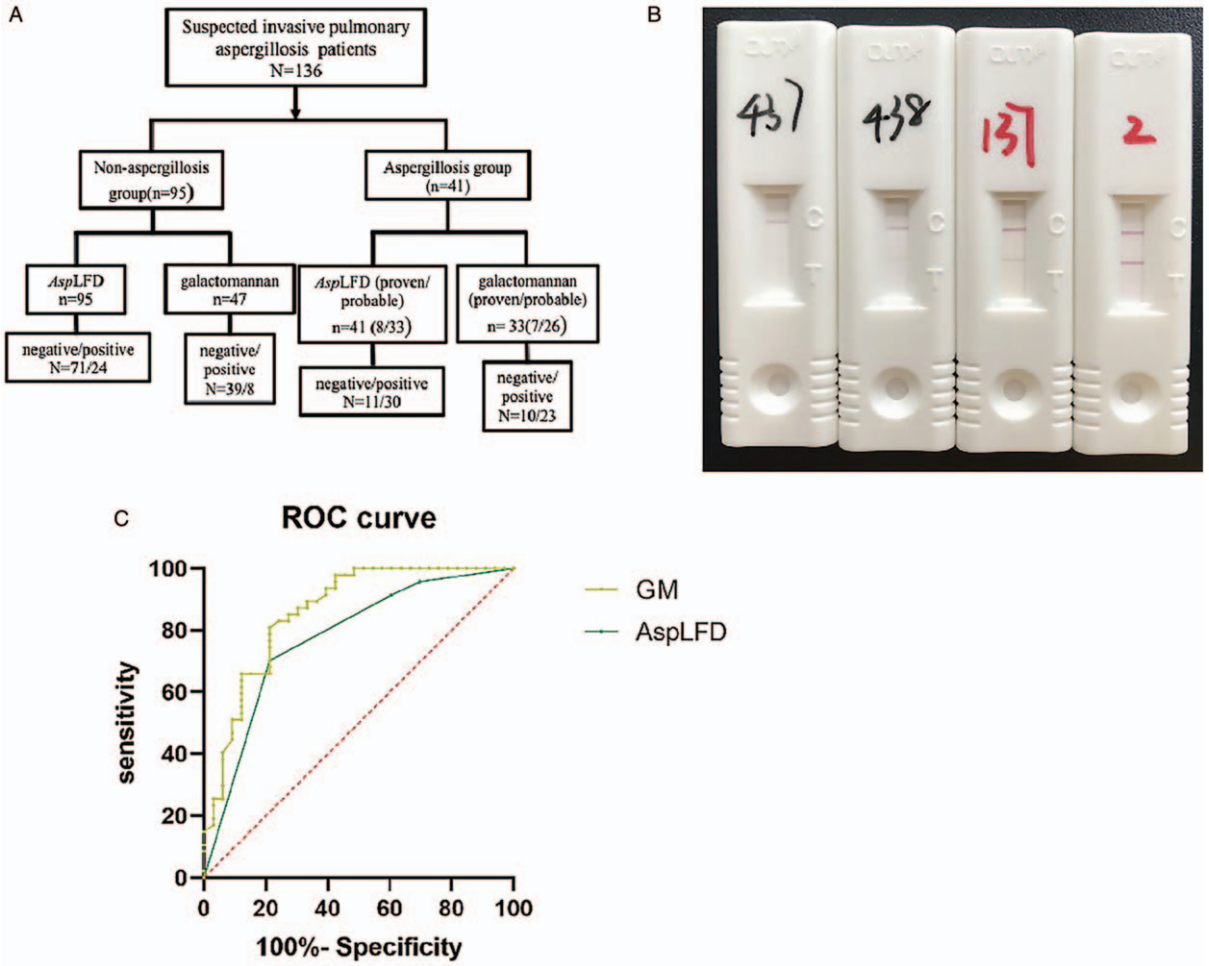

Figure 1: (A) Information of the 136 suspected non-neutropenic invasive pulmonary aspergillosis (IPA) patients; (B) Results of the *Asp*LFD using bronchoalveolar lavage fluid (BALF). Reactions typically range from negative (–, $n = 82$) to weak positive (+, $n = 32$), moderate positive (++, $n = 9$), and strong positive (+++, $n = 13$); (C) Receiver operating characteristic (ROC) curves for BALF galactomannan (GM) and *Asp*LFD test. Areas under the ROC curves: GM, 0.860 (95% confidence interval [CI]: 0.775–0.946); AspLFD, 0.775 (95% CI: 0.668–0.882).

The sensitivity, specificity, positive predictive value, negative predictive value, and diagnostic odds ratios of the *Asp*LFD for non-neutropenic IPA in all 136 BALF samples were 73.2% (30/41), 74.7% (71/95), 55.6% (30/54), 86.6% (71/82), and 8.1, respectively. The false positive rate of *Asp*LFD in patients with active tuberculosis was higher than that of other non-IPA patients (37.5% [6/16] *vs*. 22.8% [18/79]).

We compared the *Asp*LFD with the GM results in 80 patients, who received both of the test simultaneously. The results showed that the *Asp*LFD had higher sensitivity (78.8% *vs*. 69.7%, $P < 0.05$) and lower specificity (70.2% *vs*. 83.0%, $P < 0.001$) than GM. As shown in the receiver operating characteristic curve in Figure 1C, the area under the curve of GM and *Asp*LFD was 0.860 and 0.775, respectively. These results indicated that the diagnostic value of *Asp*LFD in non-neutropenic IPA patients was similar to GM.

At present, histopathology is the gold standard for IPA diagnosis. However, its application is limited. The specificity and sensitivity GM is not very high. So, combined application of multiple biomarkers is an effective way to achieve precise diagnosis.[5] Our study found that combining the positive results of the *Asp*LFD and GM lead to a higher specificity (93.6% *vs*. 83.0%, $P < 0.001$), and similar sensitivity (60.6% *vs*. 69.7%, $P = 0.1$) when compared with the GM alone. While a positive result in either the *Asp*LFD or GM or in the both lead to a higher sensitivity (90.1% *vs*. 69.7%, $P < 0.01$) and lower specificity (59.6% *vs*. 83.0%, $P = 0.001$) when compared with the GM alone. This suggested the *Asp*LFD might be useful to exclude or confirm fungal infections.

In this study, the false positive rate of the *Asp*LFD was as high as 37.5% in hospitalized patients due to tuberculosis. This false positive may be due to the increased colonization of *Aspergillus* in these patients.

In summary, we found the *Asp*LFD has clinical diagnostic value for pulmonary aspergillosis. It is quick and convenient. Therefore, the *Asp*LFD is expected to become a routine method for the clinical diagnosis of Aspergillus infection.

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Conflicts of interest

None.

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