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Mycology: Fungal infections

Diagnosis of invasive aspergillosis with two tests: ELISA and real-time PCR (MycAssay™)

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Objectives
Invasive aspergillosis (IA) has one of the highest mortality rates in patients under immunosuppressive treatment. Mainly post bone marrow or solid organ transplant patients, and patients treated for blood cancer are at high-risk. Classical microbiological diagnostics, including pathogen culture and microscopic analysis of respiratory tract samples, cannot unambiguously confirm or rule out infections. Nonculture methods, such as detecting fungal cell wall components – galactomannan antigens (GM) and 1,3-beta-D-glucan circulating in serum, and BAL, are useful in invasive aspergillosis diagnosis. MycAssay™ is the only available commercial test detecting *Aspergillus* DNA. This test was designed to detect the genomic DNA of 18 *Aspergillus* species using molecular beacon probes detecting 18S rRNA genes. The study was to assess two rapid tests in IA diagnosing – serological to detect the galactomannan antigen (ELISA assay) and Real-Time PCR, and to determine any possible correlations between these two techniques.
Methods
Nineteen patients suspected of IA, including 3 after their first kidney transplant, 5 preparing for subsequent kidney transplants, and 11 post liver transplant, were part of study. Serum samples, collected from patients during routine diagnostics, were first tested for galactomannan antigen with the ELISA assay (Platelia Aspergillus, Bio-Rad). Later, the samples, were tested with MycAssay™ (Myconostica) to establish a possible correlation between the two methods. Forty five serum samples from 19 patients suspected of IA, were tested for galactomannan. Serum samples to be tested were retrospectively selected according to their availability, IA clinical symptoms in patients, and clinical sample culture results.
Results
Serum samples from patients suspected of IA, including positive and negative galactomannan antigen, were tested. Thirty seven serum samples from 13 patients with suspected IA were tested in which no GM was detected, but in the 21 serum samples from these patients DNA were detected by Real-Time PCR (57%). In two serum samples from two patients, *Aspergillus* DNA was detected despite the lack of culture and the presence of a galactomannan. Eight serum samples obtained from seven patients with suspected IA were examined. Among these patients, in serum samples positive for galactomannan, MycAssay ™ test confirmed infection in 4 patients (50%).
Conclusions
The present study revealed both galactomannan antigen testing and the Real-Time PCR technique were useful, however, the absence of any correlation between the two methods might not confirm or rule out IA. Only the clinical symptoms, CT scan, clinical sample culture, and confirmation with one of the methods – ELISA (serological) or MycAssay™ (genetic) should be decisive. A positive MycAssay™ result in more than one serum sample and with infection symptoms, such as fever of unknown origin, abnormal CT, absence of clinical sample cultures, should be treated as potential IA. Such patients should be closely monitored and tested for invasive aspergillosis.