Galactomannan Detection as a Tool for the Diagnosis and Management of Cardiac Aspergillosis in 2 Immunocompetent Patients

Julio Garcia-Rodriguez,1 Luis Garcia-Guereta,2 Manuela De Pablos,1 Margarita Burgueros,4 and Daniel Borches3
Servicios de 1Microbiologia, 2Cardiologia Pediatrica, and 3Cirugia Cardiaca Pediatrica, Hospital Universitario La Paz, Madrid, Spain

Serum galactomannan antigen detection was used for the diagnosis and follow-up of cardiac aspergillosis after surgery in 2 nonneutropenic patients. The galactomannan index, developed in response to surgical and antifungal therapies, could prove to be a valuable method for the diagnosis and follow-up of fungal infections in such patients.

Serum galactomannan antigen detection is a useful tool for diagnosing invasive aspergillosis in selected neutropenic patients, such as hematological and bone marrow transplant recipients [1]. However, it appears to be less sensitive in other populations with immunodepression, such as patients undergoing solid-organ transplantation and critically ill patients [2, 3]. There are few reports of the value of galactomannan detection in nonneutropenic patients [3] and virtually none to support its accuracy in the diagnosis of endovascular aspergillosis, such as endocarditis or infections involving prosthetic valves or devices [4]. In this report, we present 2 cases of endovascular aspergillosis in 2 children for whom galactomannan detection proved to be a very important tool in both diagnosis and follow-up. The galactomannan antigen was measured by means of EIA (Platelia Aspergillus EIA; Bio-Rad), as specified by the manufacturer. The results were expressed in a semiquantitative mode (ratio of the optical densities of the patient samples to the mean optical density of control samples), known as the galactomannan index (GI).

The first patient was a 13-year-old nonneutropenic boy with congenital heart disease that included transposition of the great arteries, ventricular septal defect, and pulmonary stenosis. The various surgical procedures performed included the Rastelli procedure with ventricular septal defect closure and a graft connecting the right ventricle to the pulmonary artery. The boy required repeated surgical procedures because of graft stenosis. The patient experienced syncopal episodes 6 months after the last operation, after which his condition was reassessed. Echocardiography and MRI revealed severe obstruction of the pulmonary artery conduit. An additional surgical procedure was performed, and samples were sent to both the microbiology and pathology laboratories. Staphylococcus epidermidis was isolated in the cultures, but the pathology laboratory reported hyphae invading tissues. Specific treatment was initiated with liposomal amphotericin B (5 mg/kg). Three days later, treatment was supplemented by voriconazole (6 mg/kg every 12 h on the first day, followed by 4 mg/kg every 12 h). To confirm the diagnosis of invasive aspergillosis, 2 serum samples were obtained for galactomannan detection. The GIs of the first and second serum samples were 1.81 and 1.68, respectively (figure 1). Three days later, graft replacement was performed. Several samples were cultured; those cultures yielded Aspergillus nidulans. To avoid potential antagonism between antifungal drugs, voriconazole was replaced by caspofungin (70 mg/day on day 1, followed by 50 mg/day). Four days later, a GI of 1.76 was measured in another serum sample, and surgery was performed to replace the pulmonary valve and to clean the mediastinum. Samples were obtained for culture, and A. nidulans was isolated once again. Three days after the last surgery, the GI had decreased to 0.8, and a week later, the GIs were 0.45 and 0.56 in 2 consecutive samples. Two weeks later, the GI had stabilized at 0.2–0.3, values that are below the cutoff value of the assay. Cultures of blood samples were negative for Aspergillus species. One month after initiation of caspofungin treatment, the patient was discharged from the hospital while receiving oral voriconazole (4 mg/kg every 12 h) as maintenance treatment and with an order for periodic blood testing to monitor the galactomannan levels. Treatment with voriconazole was continued for 1 year. Five years later, the patient was still free of infection.

The second patient was a 12-year-old nonneutropenic boy with pulmonary valve atresia and ventricular septal defect. Surgery was performed at 3 years of age (ventricular septal defect closure and right ventricle–pulmonary artery conduit). Severe stenosis of the graft was diagnosed 8 years later; the graft was replaced with a Contegra conduit (Medtronic). The patient was discharged from the hospital in good condition and was reas-
Figure 1. Evolution of the galactomannan index (GI) in observation of patient 1 (top) and patients 2 (bottom). C, caspofungin; LA, liposomal amphotericin B; SI, surgical intervention; V, voriconazole.

sessed at periodic intervals. One year after the final surgery, he presented with fever, asthenia, and weakness. After clinical and radiological examination, a respiratory infection was diagnosed, and the patient was treated with amoxicillin for 7 days. His condition improved, but occasional febricula persisted. Three months later, he visited the emergency department with fever (temperature, 38°C) and right hypochondrial pain. Transthoracic echocardiography was performed to rule out endocarditis, and blood samples for culture were obtained. *Staphylococcus hominis* grew in 1 of 5 blood-culture bottles. Transthoracic echocardiography revealed an ill-defined mass within the graft, with moderate stenosis (gradient, 55 mm Hg). Treatment with vancomycin and gentamycin was initiated, and an MRI was performed; the MRI revealed severe intraluminal obstruction of the graft and parenchymal consolidation of the lower right segments of the lung. During the following 2 weeks, the fever resolved, but the C-reactive protein level was elevated; the patient developed marked asthenia, whereas echocardiography revealed an increase in the peak right ventricle–pulmonary artery conduit gradient of up to 100 mm Hg; fungal infection was suspected, and 2 serum samples from consecutive days were sent to the Microbiology Department for analysis of galactomannan antigen. The GIs were 5.57 and 5.36 (figure 1). Because *Aspergillus* species infection was suspected, specific treatment with caspofungin (70 mg per day followed by 50 mg per day) and voriconazole (250 mg every 12 h) was initiated. The dosages were adjusted as appropriate for the boy’s weight (65 kg). Two days later, the patient underwent an additional surgical procedure to relieve the stenosis and for graft replacement. Two specimen samples were sent to the microbiology laboratory for culture and direct examination. Septate hyphae invading tissues were detected with use of calcofluor white stain (figure 2), and *Aspergillus niger* grew in both samples. After surgical cleaning, the GI decreased to 2.5. Two serum samples per week were obtained, to monitor the response to antifungal treatment. Because of several adverse reactions, the antifungal drugs were changed, and the dosages were adjusted. Different combinations of antifungal drugs were used, including liposomal amphotericin B plus voriconazole, because our clinical experience in recent years had shown in vivo clinical success with that treatment, regardless of potential in vitro antagonism. The GI levels decreased slowly, in line with the clinical and radiological improvement (figure 1). Three months after the last hospital admission, the patient was discharged from the hospital and...
was prescribed a course of oral voriconazole (300 mg every 12 h). The most-recent follow-up echocardiography studies, performed 7 months later, revealed no obstruction of the graft. At the time of manuscript preparation (after 8 months of follow-up), the patient remained symptom free, with GI levels <0.5.

Fungal endocarditis caused by Aspergillus species is difficult to diagnose. The prognosis is poor, and diagnosis is often determined postmortem. This report describes 2 cases of Aspergillus invasive infection in nonneutropenic pediatric patients after surgical procedure(s). The sensitivity of galactomannan detection in nonneutropenic patients is very low, and experts discourage its use in this context [5]. The high rate of false-negative results has been ascribed to the presence of Aspergillus species antibodies that possibly interfere with the detection test and the rapid clearance of Aspergillus species antibodies from the blood because of renal excretion and macrophage uptake [6]. Another cause of false-negative results could be antifungal therapy, which may decrease the amount of circulating antigen [7]. Also, false-positive reactions have been described, mainly associated with gastrointestinal translocation in neonates, administration of antibiotics such as piperacillin-tazobactam or amoxicillin-clavulanate, administration of cyclophosphamide, other infections due to bacteria, and other infections due to fungi that share cross-reacting epitopes with Aspergillus species [5]. There have been other reports of endocarditis due to Aspergillus species, but serological detection of Aspergillus species did not prove to be useful for diagnosis in any of these [8]. In a previous report of Aspergillus endocarditis with a positive blood culture, the GI was negative [4]. In our patients, the galactomannan levels were high enough to confirm the clinical diagnosis that was initially suspected. This could be explained by the fact that the fungal load was greater than in former reports. Later in our study, serial measurement of GI confirmed development parallel to the clinical and radiological findings. This supports recent reports of application of the GI as a surrogate marker for evaluation of the outcome of therapy [9, 10]. For both of our patients, galactomannan levels before surgery were significantly higher than were the postoperative levels. This might be explained by a decrease in the amount of Aspergillus antigen after surgical curettage.

To our knowledge, this is the first report of the use of GI as a diagnostic tool to confirm and follow up cardiac aspergillosis involving prosthetic devices. The usefulness of determining the GI for diagnosis and follow-up in these patients should be evaluated, although a multicenter study would be essential, given the low number of cases.

Acknowledgments

Potential conflicts of interest. All authors: no conflicts.

References


Figure 2. Hyphae elements invading cardiac tissue, detected with use of calcofluor white stain.