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# High Levels of β-D-Glucan in Immunocompromised Children with Proven Invasive Fungal Disease▽

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**The plasmatic levels of 1,3-β-D-glucan (BDG) were >523 pg/ml in 4 children, 2 low-birth-weight neonates and 2 stem cell transplant recipients, with the following invasive fungal diseases (IFD) proven apart from this BDG test: 3 cases of *Candida parapsilosis* candidemias and 1 case of disseminated aspergillosis. The BDG test may be useful for identification of IFD in pediatrics.**

Invasive fungal diseases (IFD) may represent severe complications in immunocompromised children or low-birth-weight neonates. The definitions of proven and probable IFD implemented for adults (7) may also be applied successfully to immunocompromised children (4). However, while there is enough evidence of the reliability of the galactomannan antigen test for the diagnosis of invasive aspergillosis in pediatrics (5, 16, 17), no data are available for the use of 1,3-β-D-glucan (BDG) to define a diagnosis of "probable" IFD either in immunocompromised children or in low-birth-weight neonates, which represent another group at high risk of IFD (2, 3). At present, the only pediatric study available was performed with normal children using the Fungitell assay (Associates of Cape Cod, Inc., Falmouth, MA) and showed the presence of some false-positive results (15).

With the aim of evaluating the performance of the BDG test in children with proven IFD, we checked for the presence of this antigen in blood samples obtained from 4 pediatric patients with IFD already proven by positive culture from a sterile site and/or the demonstration of fungal elements in diseased tissues (7). The test was performed using the Fungitell assay (Associates of Cape Cod, Inc., Falmouth, MA), with a positive cutoff of 60 pg/ml, according to the manufacturer's recommendations. In all cases, the first positive test had to be confirmed by a second positive test performed with a sample taken more than 24 h after the first one.

Two cases were represented by low-birth-weight neonates (12 and 20 days old) with *Candida parapsilosis* candidemia, who presented values of BDG of >523 pg/ml in the presence of persistently positive blood cultures, and another case of *C. parapsilosis* candidemia with a BDG value of >523 pg/ml was observed in an 11-year-old girl receiving an allogeneic hematopoietic stem cell transplant (HSCT). The fourth case was a complex clinical condition observed in a 14-year-old boy with chronic graft-versus-host disease (GvHD), following an allogeneic HSCT. The patient was admitted into the intensive care unit for septic shock and pneumonia initially treated empirically with piperacillin-tazobactam and liposomal amphotericin B. In the following days, *Pseudomonas aeruginosa* was documented in blood and sputum cultures. The serum galactomannan antigen test initially had an index of 0.3, which increased to 0.9 in the following days, but this low positive value was attributed to piperacillin-tazobactam (12). Anyway, antibacterial treatment was shifted to meropenem for better monitoring of the risk of invasive aspergillosis, considering the possibility of concomitant infections by *P. aeruginosa* and *Aspergillus* in patients with chronic GvHD (1). In the meantime, the BDG test was also performed, and it resulted in a BDG level of 222 pg/ml. Although the blood cultures were negative, the BDG result was positive. This result was considered with suspicion because of the possibility of false-positive results in the presence of *P. aeruginosa* bacteremia (8). However, 2 days later, the patient presented a dramatic worsening of pulmonary disease, associated with the appearance of disseminated nodular skin lesions. At this time, the galactomannan antigen test had an index of 5.8, and the BDG level was >523 pg/ml. A biopsy specimen of the skin lesions showed the presence of filamentous fungi infiltrating blood vessels. Unfortunately, no pathogen grew from skin or blood cultures, and therefore, the diagnosis was that of an IFD due to a filamentous fungus, probably *Aspergillus*. Clinical data regarding all 4 patients are summarized in Table 1.

In spite of the use of different diagnostic assays, studies in neutropenic and nonneutropenic adults demonstrated that the monitoring of BDG levels might be a useful noninvasive method for the early diagnosis of IFD (6, 10, 11, 13, 14). The only pediatric data available derive from a study of normal children without IFD showing that mean glucan levels were higher in children than in adults, with a small number of false-positive results (15). Unfortunately, this study did not report any data showing the performance of BDG testing in children with IFD. Even if derived from a small number of cases, our experience shows that BDG levels may be very high in children with proven IFD. The BDG test has many causes for false-positive results, including antibiotics (amoxicillin-cla-

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TABLE 1. Demographics, underlying conditions, types and etiology, and values and timing of positive levels of 1,3-β-D-glucan in four children with documented IFD

| Patient no. | Age/sex[a] | Condition at risk | Type of documented IFD | Value (pg)/timing of first BDG serum sampling in relation to documentation of IFD |
|---|---|---|---|---|
| 1 | 12 days/M | Low-birth-wt neonate | *C. parapsilosis* candidemia | >523/same day of notification of presence of yeasts in blood culture |
| 2 | 20 days/M | Low-birth-wt neonate | *C. parapsilosis* candidemia | >523/same day of notification of presence of yeasts in blood culture |
| 3 | 11 yr/F | Allogeneic HSCT | *C. parapsilosis* candidemia | >523/same day of notification of presence of yeasts in blood culture |
| 4 | 14 yr/M | Allogeneic HSCT | Disseminated infections due to filamentous fungi, with positive galactomannan antigen | 222/3 days after documentation of pneumonia and *P. aeruginosa* bacteremia, with positive galactomannan (index of positivity, 0.9)<br>>523/3 days after first documentation of positive glucan, in the presence of skin lesions with filamentous fungi, with positive galactomannan (index of positivity, 5.8) |

[a] F, female; M, male.

vulanate) and bacterial infections (*Alcaligenes faecalis*, *Streptococcus pneumoniae*, and *P. aeruginosa*) (8, 9). We considered these possibilities, and retrospectively, this consideration led to a delay in the diagnosis for one case. We evaluated the BDG test in children with already proven IFD for exploratory purposes only; therefore, any first test (that resulted in very high levels in any case) had to be confirmed by a second one in spite of the fact that the IFD was already documented. With these assumptions, our results suggest the utility of the BDG test for immunocompromised children or low-birth-weight neonates with IFD. However, further prospective studies are needed to evaluate the right cutoff and the timing of positivity of BDG, as well as its sensitivity, specificity, and predictive values as a predictor of IFD in children at risk.

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