Group B streptococcus screening test in Obstetrical Patients

The clinical microbiology laboratory currently uses a combination of culture, molecular and immunological techniques when screening for Group B streptococcus in pre-term obstetrical patients. Following a brief, two hour incubation of the swab in broth (or overnight if the sample, is from an outside hospital), we perform a Real Time PCR (RT-PCR) assay. Following cell lysis, the bacterial DNA is amplified with PCR primers for the cfb gene (present in Group B streptococci) as well as a hybridization probe that utilizes quenched fluorophores (molecular beacons) for detection of the amplified product in real time. The RT-PCR assay takes about 1½ hours to complete.

We perform a Group B streptococcus antigen test, also following a brief incubation, on samples from patients who are admitted to the Obstetrical service during the night. The nucleic acid analysis, by RT-PCR is, however, more specific, less labor intensive, and requires less interpretation by the medical technologist than does the antigen test.

We also culture the sample overnight, after performing the PCR, and subsequently perform antibiotic susceptibility testing on a subculture. Note that the Clinical Microbiology Laboratory routinely supplements PCR with Antibiotic Susceptibility testing. PCR performed directly from a swab, does not allow this important next step. The subculture requires an additional 24 hours, following which a medical technologist selects those colonies which are group B candidates for susceptibility testing which requires another 24 hours. However, the PCR test greatly shortens the time necessary for initial detection of group B streptococci, and is highly sensitive and specific.

We recover group B strep by subsequent culture of the broth in approximately 75% of PCR positive patients. There is a significant increase in detection of positive patients utilizing this method which, in our laboratory, shows 100% sensitivity and 93% specificity when compared to culture. The molecular test, which is a significant improvement over the prior antigen test, is now the standard of care. Nevertheless we see a small percentage of samples that are PCR positive/culture negative. The consensus is that such test outcomes reflect a low level colonization, or in some instances, prior exposure to antibiotics. The decision, in such instances, whether or not to treat with antimicrobial therapy, remains one of clinical judgment.

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Serum Galactomannan Assay for Invasive Aspergillosis

Patients who are immunocompromised are at risk for a variety of infections from opportunistic organisms that rarely harm healthy individuals. Examples of these patients include oncology patients receiving chemotherapy and those on immunosuppressive therapy after bone marrow or solid organ transplant. Patients with congenital immunodeficiencies or those on chronic immunosuppressive therapy are also at risk for opportunistic infections. One type of infection that continues to be a significant cause of morbidity and mortality in these patients is invasive fungal disease. One of the most common organisms associated with fungal disease in immunocompromised patients is the environmental fungus, Aspergillus. Invasive aspergillosis is found in 5-15% of patients after allogeneic stem cell or solid organ transplant. In the past, invasive aspergillosis was diagnosed by radiographic imaging often late in the course of disease. This technique is neither sensitive nor specific for aspergillosis. Therefore, even with aggressive medical and sometimes surgical therapy outcomes were less than ideal.

The galactomannan test was developed to enhance the possibility of early diagnosis before radiologic evidence of disease. Galactomannan, a component of the polysaccharide cell wall of Aspergillus, is shed into the blood stream during invasive growth of the organism. The detection of galactomannan in the serum of at risk patients is a marker for invasive disease regardless of whether the patient has additional findings on imaging studies. It is currently being used as a screening test in the immediate post transplant period. A positive result usually leads to aggressive anti-fungal therapy and closer monitoring for other evidence of invasive Aspergillosis.

There are many studies examining the performance of the galactomannan assay in a number of different patient populations. A recent meta-analysis of 27 studies concluded that sensitivity and specificity were 0.79 and 0.89 respectively. The assay is reported to perform better in patients after stem cell transplantation than in those with solid organ transplants. However, there are fewer studies analyzing the test performance in solid organ recipients. In many studies it is reported that the galactomannan assay becomes positive 5-9 days before radiographic evidence of disease. The ability to initiate therapy approximately 1 week earlier than if only radiographic criteria were used has the potential to significantly improve outcomes. Recent data suggest that the galactomannan test can also be used to monitor the response to antifungal therapy. Levels of galactomannan fall as the infection clears and correlate with patient survival.

The Clinical Microbiology Laboratory now offers a serum galactomannan assay for the detection of invasive Aspergillus. The test employed (Platelia™ Aspergillus EIA, Bio-Rad) is a sandwich microplate ELISA. This test is recommended for patients after transplantation and oncology patients with persistent fever and neutropenia. This galactomannan assay has the best sensitivity and specificity in the allogeneic stem cell transplant population where it has been well studied and can be used in both the pediatric and adult populations. Importantly, false negative results can occur if the patient has received prior empiric antifungal therapy. Additionally, the antibiotic piperillin/tazobactam (Zosyn) administered to the patient can give a false positive serum galactomannan. The test is specific for Aspergillus and will not detect other types of invasive fungal disease such as Zygomycetes for which these patients are also at risk.

Serum galactomannan testing is performed twice weekly resulting in improved turn around times compared to refering the test to a commercial laboratory. Currently the test is primarily targeted to the population of immunocompromised patients described above, but can be performed on specimens from other patients following consultation with the laboratory medicine resident or medical microbiology fellow. For further information please phone the Microbiology Laboratory at 203-688-2460.

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