RAPID TESTS FOR DIAGNOSIS OF RESPIRATORY AND CNS VIRUSES, BACTERIAL MENINGITIS AND RESPIRATORY STREPTOCOCCAL INFECTION

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Respiratory Viral Infections
Respiratory infections due to viruses are a common cause of infections in the pediatric age group. They present with upper respiratory tract infections (URTI) such as rhinitis, pharyngitis and laryngitis and with lower respiratory tract infections (LRTI) such as bronchiolitis, bronchitis and pneumonia. The major causes of respiratory disease in children are influenza A and B viruses, parainfluenza virus (PIV) type 1 (PIV1), PIV2, PIV 3, respiratory syncytial virus (RSV), adenovirus and rhinovirus. The recent H1N1 virus also needs a special mention.

**Novel influenza A virus (H1N1)**
The Centre for Communicable Disease Control (CDC) website gives useful information on diagnostic testing for H1N1. The tests available include cell culture, direct antigen test, RNA detection by real time RT – PCR and serology (by testing acute and convalescent sera for rising antibody titres)

**Rapid influenza diagnostic tests**
Include direct antigen detection, rRT-PCR and DFA (Immunofluorescent test)

**Direct antigen test:** detects influenza viral nucleoprotein antigen. Results are available in less than 30 minutes. These tests either 1) detect and distinguish between influenza A and B virus 2) detect influenza A and influenza B virus, but cannot distinguish between them 3) detect only influenza A virus

None of these tests can distinguish seasonal influenza A virus (H3N3) from H1N1. They do not offer any information on drug susceptibility testing. Compared to r RT-PCR the sensitivity for H1N1 is 10% -70%, and for seasonal influenza virus it is 20% - 100%. The specificity is > 95%.

**r RT-PCR** – has a sensitivity between 86% - 100%.

**Direct fluorescent antibody test** has a sensitivity of 47% - 93% for H1N1 and specificity of > 96%. The test detects and distinguishes between Influenza A and B viruses, but does not distinguish between subtypes of A.

**Seasonal Influenza virus**
Isolation of the virus by tube culture may take up to two weeks. Shell vial cultures (SVC) can provide a result within 18-24 hours. The DFA test takes 2-3 hours to perform and has a sensitivity 70%-100% and a specificity of 80%-100% compared to cell culture. Rapid antigen detection tests have variable sensitivity but good specificity. They should be monitored from season to season to ensure that they pick up circulating strains. Two multiplex RT – PCR assays have been approved by the FDA. One is the Proflu+ assay, which Influenza A, Influenza B and
RSV. The second is the xTAG RVP assay, which detects 12 respiratory viruses. Molecular testing has the highest sensitivity, followed by SVC, DFA, tube culture and rapid antigen detection tests. The above mentioned tests are used for the detection of PIV1, PIV2, PIV3, RSV and Adenovirus.

Serological methods used to detect rising antibodies to respiratory viruses are not timely for diagnosis and less specific than antigen detection or molecular tests.

**Viral causes of Central Nervous System (CNS) infections**
Infections of the CNS are caused by a range of viruses. Most common among these are Herpes Simplex Virus (HSV) type 1 and type 2, Varicella-zoster virus (VZV), Enterovirus, Japanese encephalitis virus, Dengue virus, Measles, Mumps and Rubella.

**HSV 1 and 2**
These viruses can cause CNS disease as part of the primary infection or reactivation. Molecular testing has become key to the diagnosis of this infection. This test has replaced brain biopsy. Sensitivity is 100% and specificity is 94%. Even whilst the patient on acyclovir treatment the test is positive for the first week. Very early in the course of the illness, the test may be falsely negative. In patients where the clinical suspicion is high the test must be repeated 3 to 7 days into the illness. A haemorrhagic CSF sample may also give a false negative result due to inhibition of PCR. A positive CSF PCR at 10 to 14 days of treatment has led to some authorities prolonging the course of treatment. A positive PCR at the end of treatment in neonates has been shown to be a poor prognostic sign.

**VZV**
VZV can cause encephalitis as part of primary infection or reactivation. With primary infection the encephalitis occurs a week after the onset of the rash. Ataxia is the most common feature. With reactivation encephalitis can occur weeks to months after the onset of the rash. It has even been described in the absence of rash. Sensitivity of CSF PCR is variable. In primary infection it was found to be 66%, and in reactivation 44%. PCR has a higher sensitivity in children less than 10 years of age. Also if the CSF was collected within 7 days of onset of the rash the chances of detection were better.

**Enterovirus**
Enterovirus is a leading cause of aseptic meningitis, but has recently been recognized as a cause of encephalitis. Rash is a common finding. The sensitivity and specificity of RT-PCR is 100%. When faecal specimens were tested in parallel with CSF the sensitivity of faecal PCR was 96%. The sensitivity of CSF PCR decreases when obtained more than 48 hours after symptom onset. Faecal samples can be positive for more than two weeks.

**Bacterial meningitis**
CSF culture for bacteria is positive in 70-85% of patients in the absence of antibiotics for longer than 48 hours. The gram stain is positive in community acquired meningitis in 60-90% of patients. The specificity is 97%. Previous antibiotics lowers the yield by 20%. The sensitivity of
the gram stain varies with the pathogen. It is 90% for pneumococcus, 75% for meningococcus, 86% for H. influenza, 50% for other gram negative bacilli and 33% for Listeria monocytogenes.

The latex agglutination test for bacterial antigen detection is rapid and gives a result in less than 15 minutes. The sensitivity of the test is variable. 78% to 100% for H. influenza type b. 67% to 100% for pneumococcus. 69% to 100% for streptococcus group B. 50% to 93% for meningococcus. A negative antigen test does not rule out infection with a specific pathogen. Studies have shown that the result of the latex antigen test does not affect therapy in most cases. Despite having a good specificity (99.4%) false positives do occur. Hence it is felt that the test should not be done routinely. It is most useful in patients treated with antibiotics with a negative gram stain and CSF culture.

Multiplex PCRs to detect the common pathogens causing pyogenic meningitis (Meningococcus, pneumococcus and H. influenza) have shown good sensitivity and specificity.

Rapid Group A streptococcus antigen detection test (RADT)

Clinically, streptococcal pharyngitis is suspected in children with fever, tonsillar exudates, tender enlarged anterior cervical lymph nodes and absence of cough (Centor criteria). Culture of a throat swab has been the gold standard for diagnosis. This may take up to 48 hours. The rapid antigen detection test gives a result within a few minutes with a sensitivity of 70% to 85% and a specificity of 95%. The American Association of Paediatricians (AAP) recommends obtaining a follow-up throat culture in children with clinical streptococcal pharyngitis and a negative RADT. In case of a positive RADT, the diagnosis is considered positive and culture is not required. It is important to bear in mind that the sensitivity of the test varies due to multiple factors. A progressive increase in clinical criteria for streptococcal pharyngitis can cause an increase in sensitivity (from 60.9% to 95.8%) as shown in one study. Physicians who have access to laboratory tests are less likely to prescribe antibiotics than those who do not have access. The test sensitivity also depends on the person performing the test. The sensitivity is greater when laboratory personnel perform the test as opposed to non laboratory personnel.