

Hepatitis A Virus

Hepatitis A virus (HAV) is a non enveloped virus that belongs to the Hepatovirus genus in the Picornaviridae family. The viral genome consists of a linear, positive-sense, ssRNA that is approximately 7.5 kb in length. Hepatitis A is characterized as a self-limiting acute inflammatory liver disease, the symptoms of which can range from an asymptomatic form to mild illness, and even to fulminant hepatitis (<1%). HAV infection is acquired primarily by the fecal–oral route, by person-to-person contact, or by the ingestion of food or water contaminated by the feces of infected individuals.

Biochemical Diagnosis

Biochemical tests of liver function can be used as auxiliary diagnosis for viral hepatitis, and include measurement of serum total bilirubin, alkaline phosphatase, ALT and aspartate aminotransferase (AST), but only ALT is a specific test for hepatitis. In symptomatic patients, elevations of ALT and AST, alkaline phosphatase and bilirubin alkaline occurs frequently.

Serological Diagnosis

Enzyme Immunoassays (ELISA) The laboratory diagnosis of hepatitis A can be made by specific serological tests for detection of anti-HAV IgM. The presence of these antibodies in most subjects early in the infection means that their detection is the most important tool for diagnosis. Anti-HAV antibodies of the IgM class are usually detected by enzyme immunoassays (ELISA) from the onset of symptoms, and remain detectable for approximately 3 months. The anti-HAV antibodies of the IgG class are detected by total anti-HAV immunoassay, which detects IgM and IgG simultaneously.

Immunochromatographic Assay

Until now, only one immunochromatographic assay (ICA; rapid test) has been available to detect HAV antibodies. Evaluation of an ICA for the detection of anti-HAV IgM showed that the ICA showed 100% sensitivity and specificity when used to test 150 anti-HAV, IgM-positive sera collected from infected patients and 75 negative sera from healthy subjects. The enzyme immunoassays for hepatitis A are competitive assays, and detect total anti-HAV, while the rapid test detects IgM and IgG separately.

Immunohistochemical Techniques The HAV viral antigen can be detected in feces, serum, saliva, cell culture and environmental samples, usually by means of immunoassays or molecular tests. Immunohistochemical techniques utilizing direct or indirect fluorescence are generally used for identification of the HAV viral antigen in samples taken from liver, tonsils, intestine and kidneys .

Molecular Diagnosis

Molecular methods provide tools for studying HAV infection; the amplification of HAV RNA by reverse transcription, followed by PCR of the cDNA, is the most sensitive technique for screening clinical specimens. Studies using reverse transcription PCR (RT-PCR) have demonstrated that HAV RNA can be detected in blood earlier than antibodies and that the viremia may be present for a much longer period during the convalescent phase of hepatitis A than was previously thought.

Alternative Samples for Diagnosis

Dried Blood Spots

Oral Fluid

Urine Samples

Hepatitis C

Hepatitis C is an infectious disease caused by the hepatitis C virus (HCV) that primarily affects the liver. During the initial infection people often have mild or no symptoms. Occasionally a fever, dark urine, abdominal pain, and yellow tinged skin occurs. The virus persists in the liver in about 75% to 85% of those initially infected. Early on chronic infection typically has no symptoms. Over many years however, it often leads to liver disease and occasionally cirrhosis. In some cases, those with cirrhosis will develop complications such as liver failure, liver cancer, or esophageal and gastric varices .

Diagnosis of virus :

SEROLOGIC ASSAYS

Screening EIAs. The initial test used to diagnose HCV is an enzyme immunoassay (EIA) for anti-HCV immunoglobulin G (IgG). The HCV genome encodes a polyprotein of 3,011 to 3,033 amino acids that is processed into 10 structural and nonstructural (NS) proteins.

Enzyme Immunoassays (ELISA) The laboratory diagnosis

Use of serologic assays

Laboratories detect HCV RNA with commercially available assay kits of signal amplification step is needed. Reverse transcriptase (RT) PCR (RT-PCR) and transcription-mediated amplification (TMA) are target amplification methods. The branched DNA (bDNA) assay is a signal amplification technique.

Rubella

Rubella, also known as German measles or three-day measles, is an infection caused by the rubella virus. This disease is often mild with half of people not realizing that they are sick. A rash may start around two weeks after exposure and last for three days. It usually starts on the face and spreads to the rest of the body. The rash is not as bright as that of measles and is sometimes itchy. Swollen lymph nodes are common and may last a few weeks. A fever, sore throat, and fatigue may also occur. In adults joint pain is common. Complications may include bleeding problems, testicular swelling, and inflammation of nerves. Infection during early pregnancy may result in a child born with congenital rubella syndrome (CRS) or miscarriage. Symptoms of CRS include problems with the eyes such as cataracts, ears such as deafness, heart, and brain. Problems are rare after the 20th week of pregnancy.



Laboratory Diagnosis

A. Serological diagnosis of rubella infection - Serology is the mainstay of diagnosis of rubella infection. A recent rubella infection can be diagnosed by (1) detection of rubella-specific IgM, (2) rising titres of antibody in HAI and

ELISA tests, and (3) seroconversion. Blood should be collected from pregnant women with features of rubella-like illness as soon as possible after onset of symptoms.

1. Haemagglutination inhibition (HAI) assay remains the mainstay test for diagnosis of rubella infection. HAI Abs may be detected on the first day of the rash and rise rapidly to peak titres. It may be difficult to demonstrate a significant rise in titre unless the serum is obtained within the first few days of the onset of illness. Before testing by HAI, sera must be treated to remove red cell agglutinins and serum lipoproteins. It is current practice to confirm a serological diagnosis by HI with testing for rubella IgM.

2. EIA and RIA have replaced HAI for the diagnosis of rubella in some laboratories. Occasionally SRH can be used for diagnostic purposes provided the wells are of consistent size and a measured volume of serum is added to each well.

3. Detection of rubella-specific IgM by EIA or RIA. The most sensitive and reliable techniques in use are tM - antibody capture ELISA and radioimmunoassay). IgM antiglobulins such as Rheumatoid Factor can seldom cause false positive results as can heterophil antibodies. Indirect ELISA and RIA has also been developed but these are not as sensitive as direct ELISA .

B. Molecular diagnosis by PCR .