IM LATEX TEST KIT

Latex agglutination slide test for the qualitative and semi-quantitative determination of infectious mononucleosis in non-diluted serum or plasma.

INTRODUCTION

Infectious mononucleosis involves the reticuloendothelial tissue and is believed to be caused by the Epstein Barr virus. It is generally limited to and affects children and young adults. Infectious Mononucleosis may be confused on a symptomatic basis with other diseases. For this reason, an accurate diagnosis using this procedure is necessary.

Detectable levels of unique heterophile antibodies are produced in patients with infectious mononucleosis[2]. The antibody of the IM was shown by Paul and Bunnell [2] to agglutinate sheep and horse erythrocytes and Bunnell [3] subsequently attempted to use this observation as a basis for screening. A specific test was not developed until Davidsohn[4567] modified the procedure by introducing differntial absorption steps to eliminate Forssman and serum sickness antibody confusion. The Davidsohn test procedure is accepted as the classic reference method in detecting IM.

The IM latex test provides a suspension of polystyrene latex particles which have been coated with partially purified glycoprotein from bovine red blood cells. The heterophile antibody associated with IM blinds to the corresponding antigenic determinants on the glycoprotein coated latex. Due to the purification of the bovine red blood cell, the glycoprotein coated latex is not agglutinated by Forssman or serum sickness antibodies at levels normally encountered in the U.K. population; therefore, no differential absorption is required.

Utilising our test serum found positive for IM which contains heterophile antibody associated with IM binds to the corresponding antigenic determinants on the glycoprotein coated latex forming visible macroagglutination.

STANDARD KIT PRESENTATION

1. Latex reagent sufficient for 50/100 slide tests. Suspension of polystyrene latex particles coated with partially purified glycoprotien from bovine red cells.
2. Positive Control. Stablilized human serum, known to have a positive reaction with the IM latex reagent.
3. Negative Control. Stabilized human serum, known to have a negative reaction with the IM latex reagent.
4. Reusable plastic test slide.
5. Disposable pipette/mixers.

WARNINGS AND PRECAUTIONS

For In Vitro Diagnostic Use. The controls used in this kit have been tested by an FDA approved method and found non-reactive for the presence of HbsAg and antibody to HIV. While these methods are highly accurate, no test can offer complete assurance that infectious agents are absent. This material, as well as all patient samples, should be handled as though capable of transmitting infectious disease.

The United States Food and Drug Administration recommends such samples be handled as Center for Disease Control's Biosafety Level 2.

Reagents in this kit contain sodium azide as a perservative. Sodium azide has been reported to form lead or copper azide in laboratory plumbing which may explode on percussion. Flush drains with water thoroughly after disposing of fluids containing sodium azide.

STORAGE

Reagents are stable when stored at 2-8°C until the expiration date shown on their respective labels. Lack of clear agglutination with the positive control or extreme turbidity in either control serum indicates deterioration.

DO NOT FREEZE!

ADDITIONAL REQUIREMENTS

Timer
Slide Rotator (optional)
Light Source
Physiological saline (0.85% or 0.9% sodium chloride)
Test Tubes & Rack (Semi-quantitative test only)
Serological Pipettes (Semi-quantitative test only)

SAMPLE PREPARATION

It is recommended that serum or plasma free from contamination be used. Serum samples must have clotted completely and contain no particulates nor traces of fibrin after thawing. Do not heat inactivate test sera or controls. Avoid repeated freeze/thawing of specimens. Serum specimens are reportedly stable up to 2 days at 2-8°C and for up to 2 weeks if they are frozen (at or below -20°C). As in all serological tests, hemolytic, lipemic or turbid sera may cause incorrect results and should not be used. Use only a clean, dry slide washed in mild detergent and rinsed thoroughly with distilled water.

LIMITATIONS

Reaction times greater than 3 minutes can lead to false positive results (due to drying effect). Very lipemic sera can also cause non-specific reactions.

Although the IM Test is a highly sensitive and specific, a diagnosis of infectious mononucleosis should not be made on the basis of a positive test result without the support of patient history and haematological or other clinical evidence. Similarly, a negative result cannot completely rule out infectious mononucleosis.

Apparent false positive reactions have been associated with sera from patients with other diseases such as infections, leukemia, Burkitt's lymphoma and serum sickness [8,9,10,11,12].

IM LATEX TEST KIT

Although most patients develop heterophile antibodies within 3 weeks of the onset of symptoms, occasional patients may take up to several months to develop detectable levels. If the IM Test is negative in the presence of strong evidence suggesting a diagnosis of infectious mononucleosis, repeat testing on samples obtained at intervals of several days will generally reveal development of the heterophile agglutinin. Some patients with haematological and clinical evidence of IM remain persistently negative [12,13,14].

A single heterophile antibody titre cannot be interpreted as an indication of the stage or severity of the disease [15,16]. However, titrations on sequential samples may be useful in following the course of the disease in an individual patient.

QUALITATIVE METHOD

1. Bring reagents and patient samples to room temperature
2. Gently mix the latex reagent vial to disperse and suspend the latex particles. Do Not Shake Vigorously!
3. Using a dispensing pipette provided, add one drop (40µL) of the non-diluted patient samples(s) onto a separate cell(s) on the slide. Also place one (1) drop of the positive and negative control on separate cells of the test slide.
4. Gently mix the contents of the latex reagent and place one (1) drop next to the drop of serum sample(s) on each of the separate cells in use.
5. Mix both drops with the mixer provided, covering the whole surface of each of the cell(s).
6. Gently rotate the slide for 3 minutes manually or on a rotary shaker set at 80 – 100 rpm.
7. After the 3 minutes, examine each cell(s) for the absence or presence of agglutination.

A positive control and a negative control should be included in each test series. A positive control will produce a coarse agglutination against a clear background, while a negative control will produce a smooth, homogeneous suspension.

SEMI-QUANTITATIVE METHOD

1.This reaction can be used to estimate the IM titre using a dilution series. The patient sample is diluted with physiological saline as shown:

Dilution			
1:2	(1 part serum	+	1 part saline)
1:4	(1 part serum	+	3 parts saline)
1:8	(1 part serum	+	7 parts saline)
1:16	(1 part serum	+	15 parts saline)

Thereafter, each is tested with the latex as described under Procedure section "A. Qualitative Method".

QUALITATIVE METHOD RESULTS

Agglutination identifies the presence of the heterophile antibody in the sample. Samples that do not show agglutination indicate the absence of the heterophile agglutinin associated with infectious mononucleosis.

SEMI-QUANTATIVE METHOD RESULTS

The titre of IM heterophile antibody is the reciprocal of the highest dilution which exhibits a positive result. The actual titre of the antibody has not been related to the stage or severity of the disease [15,16]. However, an increase in the IM heterophile agglutination titre may be clinically significant in the early stages of the disease and may assist in the diagnosis of IM.

EXPECTED VALUES

Detectable levels of the IM heterophile antibody can usually be expected to occur between the 6th and 10th day following the onset of symptoms. The level usually increases through the second or third week of illness and, thereafter, can be expected to persist with gradual decline over a 12 month period. Positive results should be seen in approximately 98% of all IM cases.[17]

PERFORMANCE CHARACTERISTICS

Correlation:Serum and plasma specimens from 285 individuals which had been submitted to clinical laboratories by physicians for IM testing were examined. The IM Latex Test and a commercial RBC kit were used to evaluate the specimens. 132 specimens were found to be positive using both assays. The remaining 153 specimens gave negative results using both assays. Therefore this data indicates that both sensitivity and specificity of the IM Latex Test are 100%

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