PLATELIA™ HSV 1+2 IgM

96 TESTS

QUALITATIVE DETECTION OF IgM ANTIBODIES TO HSV 1 AND HSV 2 IN HUMAN SERUM OR PLASMA BY ENZYME IMMUNOASSAY

IVD
1. INTENDED USE

Platelia™ HSV 1+2 IgM is an immunoassay using immunocapture format for qualitative detection of IgM antibodies to Herpes simplex viruses from type 1 and type 2 in human serum or plasma.

2. CLINICAL VALUE

Herpes simplex virus (HSV) is a common human pathogen found worldwide. Two serological subtypes of HSV have been described: HSV-1 and HSV-2. HSV-1 is primarily associated with infection in the tongue, mouth, lips, pharynx and eyes; whereas HSV-2 is primarily associated with genital and neonatal infection. However, both types can cause genital infections, and HSV-1 is increasingly recognized as a cause of genital symptoms.

Herpes simplex virus is transmitted through direct contact with secretions from an infected person, who may or may not have disease symptoms at the time. In fact, most genital infections are transmitted in the absence of symptoms. Primary infection with HSV can occur at any age, affecting neonates, children, and adults. Following primary infection, HSV establishes lifelong latency in sensory nerve ganglia, causing subsequent recurrence of symptoms when latent virus is reactivated.

Expectant mothers who acquire HSV type-1 or type-2 during pregnancy can transmit the virus to the infant before birth or during delivery. In utero infections are associated with spontaneous abortion and premature delivery. The greatest risk of neonatal herpes is to babies whose mother’s contract genital infection in the final trimester of pregnancy. Congenital and neonatal HSV can occur with primary or recurrent, symptomatic or asymptomatic, maternal HSV infection and can cause skin, eye or mouth infections, and damage to the central nervous system.

In primary HSV infections, IgM antibodies usually appear between the third and seventh day after onset of symptoms. IgM antibody titer peaks in four to six weeks and usually decline to undetectable levels after two months. IgM antibodies to HSV can sometimes be found in recurrent infections. However, production and detection of anti-HSV-IgM antibodies in patients with recurrent infections is less predictable and may be related to the severity of infection. IgG antibodies to HSV usually appear one to two weeks after the onset of infection and persist at various levels for life. Presence of anti-HSV IgG antibodies cannot discriminate between recent infection and prior exposure (latent infection or reactivation) to Herpes simplex virus. In such situation, residual IgM can be observed and detection of anti-HSV IgM antibodies may not be relevant. However, detection of anti-HSV IgM antibodies is clinically important during early acute infection when anti-HSV IgG antibodies could not be present yet.(2)
3. **PRINCIPLE**

Platelia™ HSV 1+2 IgM is a qualitative test for detection of IgM antibodies to HSV-1 and HSV-2 in human serum or plasma by enzyme immunoassay with capture of the IgM on the solid phase.

Anti-human µ-chains antibodies are coated on the solid phase (wells of the microplate). An HSV antigen labeled with peroxidase is used as the conjugate. The test uses the following steps:

- **Step 1**
  Patients samples, calibrator and controls are diluted 1/21 and then distributed in the wells of the microplate. During this incubation of one hour at 37°C, IgM antibodies present in the sample bind to the anti-µ antibodies coated on the microplate wells. After incubation, IgG and other serum proteins are removed by washings.

- **Step 2**
  The conjugate (HSV antigen labeled with peroxidase) is added to the microplate wells. During this incubation of one hour at 37°C, the conjugate binds to the specific IgM anti-HSV antibodies that were eventually captured on the microplate. The unbound conjugate is removed by washings at the end of the incubation.

- **Step 3**
  The presence of immune-complexes (Anti-human µ-chains / IgM anti-HSV / HSV Antigen labeled with peroxidase) is demonstrated by the addition in each well of an enzymatic development solution.

- **Step 4**
  After incubation at room temperature (+18-30°C), the enzymatic reaction is stopped by addition of 1N sulfuric acid solution. The optical density reading obtained with a spectrophotometer set at 450/620 nm is proportional to the amount of IgM antibodies to HSV present in the sample.
4. **PRODUCT INFORMATION**

Supplied quantities of reagents have been calculated to allow 96 tests. All reagents are exclusively for *in vitro* diagnostic use.

<table>
<thead>
<tr>
<th>Label</th>
<th>Nature of reagents</th>
<th>Presentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>R1</td>
<td>Microplate (Ready-to-use): 12 strips with 8 breakable wells, coated with anti-human μ chains</td>
<td>1</td>
</tr>
<tr>
<td>R2</td>
<td>Concentrated Washing Solution (20x): TRIS-NaCl buffer (pH 7.4), 2% Tween® 20 Preservative : &lt; 1.5% ProClin™ 300</td>
<td>1 x 70 mL</td>
</tr>
<tr>
<td>R3</td>
<td>Negative Control: Human serum negative for IgM antibodies to HSV, and negative for HBs antigen, anti-HIV1, anti- HIV2 and anti-HCV Preservative : &lt; 1.5% ProClin™ 300</td>
<td>1 x 0.75 ml</td>
</tr>
<tr>
<td>R4</td>
<td>Calibrator: Human serum reactive for IgM antibodies to HSV, and negative for HBs antigen, anti-HIV1, anti- HIV2 and anti-HCV Preservative : &lt; 1.5% ProClin™ 300</td>
<td>1 x 0.75 ml</td>
</tr>
<tr>
<td>R5</td>
<td>Positive Control: Human serum reactive for IgM antibodies to HSV, and negative for HBs antigen, anti-HIV1, anti- HIV2 and anti-HCV Preservative : &lt; 1.5% ProClin™ 300</td>
<td>1 x 0.75 ml</td>
</tr>
<tr>
<td>R6</td>
<td>Conjugate (Ready-to-use): Viral antigen lysate and gG1 HSV-1 protein labeled with peroxydase Preservative : &lt; 1.5% ProClin™ 300</td>
<td>2 x 13 ml</td>
</tr>
<tr>
<td>R7</td>
<td>Diluent for samples (Ready-to-use): TRIS-NaCl (pH 8.5), milk, phenol red Preservative : &lt; 1.5% ProClin™ 300</td>
<td>1 x 40 ml</td>
</tr>
</tbody>
</table>
5. WARNINGS AND PRECAUTIONS

The reliability of the results depends on correct implementation of the following Good Laboratory Practices:

- Do not use expired reagents.
- Do not mix or associate within a given run reagents from different lots.

**REMARK:** For Washing Solution (R2, label identification: 20x colored green), Chromogen (R9, label identification: TMB colored turquoise) and Stopping Solution (R10, label identification: 1N colored red), it is possible to use other lots than those contained in the kit, provided these reagents are strictly equivalent and the same lot is used within a given test run.

**REMARK:** In addition, the Washing Solution (R2, label identification: 20x colored green) can be mixed with the 2 other washing solutions included in various Bio-Rad reagent kits (R2, label identifications: 10x colored blue or 10x colored orange) when properly reconstituted, provided only one mixture is used within a given test run.

- Before use, wait for 30 minutes to allow reagents to reach room temperature (+18-30°C).
- Carefully reconstitute or dilute the reagents avoiding any contamination.
- Do not carry out the test in the presence of reactive vapors (acid, alkaline, aldehyde vapors) or dust that could alter the enzyme activity of the conjugate.
- Use glassware thoroughly washed and rinsed with deionized water or, preferably disposable material.
- Washing the microplate is a critical step in the procedure: follow the recommended number of washings cycles and make sure that all wells are completely filled and then completely emptied. Incorrect washings may lead to inaccurate results.

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<table>
<thead>
<tr>
<th>Label</th>
<th>Nature of reagents</th>
<th>Presentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>R9</td>
<td>Chromogen TMB</td>
<td>Chromogen (Ready-to-use): 3.3’5.5’ tetramethylbenzidine (&lt; 0.1%), H₂O₂ (&lt; 1%)</td>
</tr>
<tr>
<td>R10</td>
<td>Stopping Solution</td>
<td>Stopping Solution (Ready-to-use): 1N sulfuric acid solution</td>
</tr>
<tr>
<td></td>
<td>Plate sealers</td>
<td>4</td>
</tr>
</tbody>
</table>

For storage conditions and expiration date, please refer to the indications stated on the box.
• Do not allow the microplate to dry between the end of the washings operation and the reagent distribution.
• Never use the same container to distribute the conjugate and the development solution.

**REMARK:** The conjugate being particularly concentrated in peroxidase, any contact with the TMB solution (droplet, spilling) could lead to false positive reaction. Avoid distributing the conjugate near any material (tips, racks) that will be used for the TMB distribution step.

• The enzymatic reaction is very sensitive to metal or metal ions. Consequently, do not allow any metal element to come into contact with the various solutions containing the conjugate or the chromogen.
• Chromogen solution (R9) should be colorless. The appearance of a blue color indicates that the reagent cannot be used and must be replaced.
• Use a new pipette tip for each sample.
• Check the pipettes and other equipments for accuracy and correct operations.

**HEALTH AND SAFETY INSTRUCTIONS**

Human origin material used in the preparation of reagents has been tested and found non-reactive for hepatitis B surface antigen (HBs Ag), antibodies for hepatitis C virus (anti-HCV), and to human immunodeficiency virus (anti-HIV1 and anti-HIV2). Because no method can absolutely guarantee the absence of infectious agents, handle reagents of human origin and patient samples as potentially capable of transmitting infectious diseases:

• Any material, including washings solutions, that comes directly in contact with samples and reagents containing materials of human origin should be considered capable of transmitting infectious diseases.
• Wear disposable gloves when handling samples and reagents.
• Do not pipette by mouth.
• Avoid spilling samples or solutions containing samples. Spills must be rinsed with bleach diluted to 10%. In the event of a spill with an acid, it must be first neutralized with sodium bicarbonate, and then cleaned with bleach diluted to 10% and dried with adsorbent paper. The material used for cleaning must be discarded in a contaminated residue container.
• Patient samples, reagents containing human origin material, as well as contaminated material and products should be discarded after decontamination only:
  - either by immersion in bleach at the final concentration of 5% of sodium hypochloride during 30 minutes,
  - or by autoclaving at 121°C for 2 hours at the minimum.
CAUTION: Do not introduce solutions containing sodium hypochloride into the autoclave

- Avoid any contact of reagents, including those considered as not dangerous, with skin and mucosa.
- Chemical and biological residues must be handled and disposed off in accordance with Good Laboratories Practices.
- All reagents in the kit are exclusively for in vitro diagnostic use.

Caution: Some of the reagents contain Proclin™ 300 < 1.5%
R43: May cause sensitisation by skin contact
S28-37: After contact with skin, wash immediately with plenty of water and soap. Wear suitable gloves

6. SAMPLES

1. Serum and plasma (EDTA, heparin or citrate) are the recommended sample types.
2. Observe the following recommendations for handling, processing and storage of blood samples:
   - Collect all blood samples observing routine precaution for venipuncture.
   - For serum, allow samples to clot completely before centrifugation.
   - Keep tubes stoppered at all times.
   - After centrifugation, separate the serum or plasma from the clot or red cells in a tightly stoppered storage tube.
   - The specimens can be stored at +2-8°C if test is performed within 7 days.
   - If test will not be completed within 7 days, or for shipment, freeze the samples at -20°C or colder.
   - Do not use samples that have been thawed more than five times. Previously frozen specimens should be thoroughly mixed (Vortex) after thawing prior to testing.
3. Samples containing 90 g/l of albumin or 100 mg/l of unconjugated bilirubin, lipemic samples containing the equivalent of 36 g/l of triolein (triglyceride), and hemolysed samples containing up to 10 g/l of haemoglobin do not affect the results.
4. Do not heat the samples.

7. ASSAY PROCEDURE

7.1 MATERIALS REQUIRED BUT NOT PROVIDED

- Vortex mixer.
- Microplate reader equipped with 450 nm and 620 nm filters (*).
- Microplate incubator thermostatically set at 37±1°C (*).
- Automatic, semi-automatic or manual microplate washer (*).
• Sterile distilled or deionized water.
• Disposable gloves.
• Goggles or safety glasses.
• Adsorbent paper.
• Automatic or semi-automatic, adjustable or preset, pipettes or multi-pipettes, to measure and dispense 10 µl to 1000 µl, and 1 ml, 2 ml and 10 ml.
• Graduated cylinders of 25 ml, 50 ml, 100 ml and 1000 ml capacity.
• Sodium hypochloride (bleach) and sodium bicarbonate.
• Container for biohazard waste.
• Disposable tubes.
(*)Consult our technical department for detailed information about the recommended equipment.

7.2 REAGENTS RECONSTITUTION
• **R1:** Allow 30 minutes at room temperature (+18-30°C) before opening the bag. Take out the carrier tray, return unused strips in the bag immediately and check the presence of desiccant. Carefully reseal the bag and store it at +2-8°C.
• **R2:** Dilute 1/20 the washing solution R2 in distilled water: for example 50 ml of R2 and 950 ml of distilled water to get the ready-to-use washing solution. Prepare 350 ml of diluted washing solution for one plate of 12 strips if washing manually.
• **R3, R4, R5:** Dilute 1/21 in Diluent (R7) (example: 300 µl of R7 + 15 µL of Calibrator or Control).

7.3 STORAGE AND VALIDITY OF OPENED AND / OR RECONSTITUTED REAGENTS
The kit must be stored at +2-8°C. When the kit is stored at +2-8°C before opening, each component can be used until the expiration date indicated on the outer label of the kit.
• **R1:** Once opened, the strips remain stable for up to 8 weeks if stored at +2-8°C in the same carefully closed bag (check the presence of desiccant).
• **R2:** Once diluted, the Washing Solution can be kept for 2 weeks at +2-30°C. Once opened, the concentrated Washing Solution stored at +2-30°C, in absence of contamination, is stable until the expiration date indicated on the label.
• **R9:** Once opened and without any contamination, the reagent stored at +2-8°C is stable for up to 8 weeks.
• **R10:** Once opened and without any contamination, the reagent stored at +2-8°C is stable until the expiration date indicated on the label.
7.4 PROCEDURE

Strictly follow the assay procedure and Good Laboratory Practices.
Before use, allow reagents to reach room temperature (+18-30°C).
The use of breakable wells requires a special attention during handling.
Use calibrator and controls with each run to validate the assay results.

1. Carefully establish the distribution and identification plan for calibrator, controls and patients samples.
2. Prepare the diluted Washing Solution (R2) [Refer to Section 7.2].
3. Take the carrier tray and the strips (R1) out of the protective pouch [Refer to Section 7.2].
4. In individually identified tubes, dilute Calibrator and Controls (R3, R4, R5) and patients samples (S1, S2...) in Diluent (R7) to give a 1/21 dilution: 300 µl of Diluent (R7) and 15 µl of sample. Vortex diluted samples.
5. Strictly following the indicated sequence below, distribute in each well with 200µl of diluted calibrator, controls and patient samples:

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<th>10</th>
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<th>12</th>
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<tbody>
<tr>
<td>A</td>
<td>R3</td>
<td>S5</td>
<td>S13</td>
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<td>G</td>
<td>S3</td>
<td>S11</td>
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<td>H</td>
<td>S4</td>
<td>S12</td>
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</tbody>
</table>

6. **Cover the microplate with an adhesive** plate sealer, then press firmly onto the plate to ensure a tight seal. Incubate the microplate immediately in a thermostat controlled water bath or in a dry incubator for 1 hour ± 5 minutes at 37°C ± 1°C.

7. At the end of the first incubation period, remove the adhesive plate sealer. Aspirate the content of all wells into a container for biohazard waste (containing sodium hypochloride). Wash microplate 5 times with 350 µl of the Washing Solution (R2). Invert the microplate and gently tap on adsorbent paper to remove remaining liquid.

8. Distribute immediately 200 µl of the conjugate (R6) in all wells [Warning-Refer to Section 5].
9. **Cover the microplate with an adhesive plate sealer**, then press firmly onto the plate to ensure a tight seal. Incubate the microplate immediately in a thermostat controlled water bath or in a dry incubator for 1 hour ± 5 minutes at 37°C ± 1°C.

10. At the end of the second incubation period, remove the adhesive plate sealer. Aspirate the content of all wells into a container for biohazard waste (containing sodium hypochloride). Wash microplate **5 times** with 350 µl of the Washing Solution (R2). Invert the microplate and gently tap on adsorbent paper to remove remaining liquid.

11. Quickly distribute into each well and **away from light** 200 µl of Chromogen solution (R9). **Allow the reaction to develop in the dark for 30 ± 5 minutes at room temperature (+18-30°C)**. Do no use adhesive plate sealer during this incubation.

12. Stop the enzymatic reaction by adding 100 µl of Stopping Solution (R10) in each well. Use the same sequence and rate of distribution as for the development solution.

13. Carefully wipe the plate bottom. Read the optical density at 450/620 nm using a plate reader within 30 minutes after stopping the reaction. The strips must always be kept away from light before reading.

14. Before reporting results, check for agreement between the reading and the distribution plan of plate and samples.

### 8. INTERPRETATION OF RESULTS

#### 8.1 CALCULATION OF THE CUT-OFF VALUE (CO)

The Cut-Off value (CO) corresponds to the mean value of the optical densities (OD) of the cut-off Control duplicates (R4):

- \( \text{CO} = \text{mean of OD R4} \)

#### 8.2 CALCULATION OF THE SAMPLE RATIO

Sample result is expressed by Ratio using the following formula:

- Sample Ratio = Sample OD/CO

#### 8.3 QUALITY CONTROL

Include the calibrator and controls for each microplate and for each run, and analyse the obtained results. For validation of the assay, the following criteria must be met:

- Optical density values:
  - \( \text{CO} \geq 0.200 \)
  - \( 0.80 \times \text{CO} < \text{OD R4 Repl.1} < 1.20 \times \text{CO} \)
  - \( 0.80 \times \text{CO} < \text{OD R4 Repl.2} < 1.20 \times \text{CO} \)

(Individual OD of each replicate of the Cut-Off control (R4) must not differ more than 20% of the CO value).
- Optical density ratios:
  - Ratio R3 (OD R3 / CO) ≤ 0.60
  - Ratio R5 (OD R5 / CO) ≥ 1.50

If those quality control criteria are not met, the test run should be repeated.

**8.4 INTERPRETATION OF RESULTS**

<table>
<thead>
<tr>
<th>Sample Ratio</th>
<th>Result</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ratio &lt; 0.90</td>
<td>Negative</td>
<td>The sample is considered non reactive for the presence of IgM antibodies to HSV-1 and HSV-2</td>
</tr>
<tr>
<td>0.90 ≤ Ratio &lt; 1.10</td>
<td>Equivocal</td>
<td>The sample is considered equivocal for the presence of IgM antibodies to HSV-1 and/or HSV-2. The result must be confirmed by another test done on a second sample.</td>
</tr>
<tr>
<td>Ratio ≥ 1.10</td>
<td>Positive</td>
<td>The sample is considered reactive for the presence of IgM antibodies to HSV-1 and/or HSV-2.</td>
</tr>
</tbody>
</table>

**8.5 TROUBLE SHOOTING GUIDE**

Non validated or non repeatable reactions are often caused by:
- Inadequate microplate washings.
- Contamination of negative samples by serum or plasma with a high antibody titer.
- Contamination of the development solution by chemical oxidizing agents (bleach, metal ions...).
- Contamination of the Stopping Solution.

**9. PERFORMANCES**

Platelia™ HSV 1+2 IgM was evaluated on 2 different sites on a total of 595 samples. Results with Platelia™ HSV 1+2 IgM were compared to results obtained with an other commercialized EIA assay.

**9.1 PREVALENCE**

Prevalence determination of IgM antibodies to Herpes Simplex Virus Type 1+2 in human serum was estimated using a panel of 89 samples obtained from pregnant women. The following results were obtained: 66 negative, 7 doubtful and 16 positive sera. Prevalence using the Platelia™ HSV 1+2 IgM assay is established at 18.0% (16/89).

**9.2 RELATIVE SPECIFICITY**

Specificity was estimated on a total of 480 samples:
- on site 1, a panel of 233 samples from blood donor, pregnant women, HSV serology requests and commercial panel (BBI®).
- on site 2, a panel of 247 samples from hospitalized patients.

On both sites, samples were selected on negative results obtained with a commercialized IgM EIA assay and considered as a reference.
It is to be noticed that, among the 60 sera found doubtful or positive with Platelia™ HSV 1+2 IgM, 47 were found positive with Platelia™ HSV 1 IgG assay and/or Platelia™ HSV 2 IgG.

In addition, one BBI panel serum found positive with Platelia™ HSV 1+2 IgM assay and negative with the commercialized IgM EIA and Platelia™ HSV 1 IgG and Platelia™ HSV 2 IgG was found positive with 6 different IgM assays.

### 9.3 RELATIVE SENSITIVITY

Sensitivity was estimated on a total of 87 samples:

- on site 1, a panel of 71 samples from blood donor, pregnant women, HSV serology requests and commercial panel (BBI\(^{(a)}\)).
- on site 2, a panel of 16 samples from hospitalized patients.

On both sites, samples were selected on positive results obtained with a commercialized IgM EIA assay and considered as a reference.

<table>
<thead>
<tr>
<th>Panel of samples</th>
<th>Negative</th>
<th>Doubtful (^{(1)})</th>
<th>Positive</th>
<th>Relative Sensitivity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Site 1</td>
<td>N = 71</td>
<td>5</td>
<td>0</td>
<td>93.0% (66/71)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>[84.3%-97.7%]</td>
</tr>
<tr>
<td>Site 2</td>
<td>N = 16</td>
<td>2</td>
<td>1</td>
<td>86.7% (13/15)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>[59.5%-98.3%]</td>
</tr>
<tr>
<td>Total</td>
<td>N = 87</td>
<td>7</td>
<td>1</td>
<td>91.9% (79/86)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
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<td>[84.0%-96.7%]</td>
</tr>
</tbody>
</table>

\(^{(1)}\) doubtful results were excluded for calculation of sensitivity
It is to be noticed that, among the 7 sera found negative with Platelia™ HSV 1+2 IgM, 3 were found positive with Platelia™ HSV 1 IgG (72820) and/or Platelia™ HSV 2 IgG (72821).

9.4 SEROCONVERSIONS
Among a panel of 33 patients presenting a seroconversion, 22 were simultaneously found positive or doubtfull with Platelia™ HSV 1+2 IgM assay and the other commercialized IgM EIA assay, 7 were first detected by Platelia™ HSV 1+2 IgM assay, 3 were found positive only with the other commercialized IgM EIA assay and 1 serum was not detected by neither assay.

9.5 PRECISION
- Within-run precision (repeatability):
In order to evaluate intra-assay repeatability, one negative, one equivocal and one positive samples were tested 30 times during the same run. The ratio (Sample OD / CO) was determined for each sample. Mean Ratio, Standard Deviation (SD) and Coefficient of Variation (%CV) for each of the three specimens are listed in table below:

<table>
<thead>
<tr>
<th>N=30</th>
<th>Negative Sample</th>
<th>Low Positive sample</th>
<th>High Positive sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ratio (Sample OD / Cut-Off value)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>0.32</td>
<td>2.22</td>
<td>4.06</td>
</tr>
<tr>
<td>SD</td>
<td>0.01</td>
<td>0.20</td>
<td>0.10</td>
</tr>
<tr>
<td>% CV</td>
<td>2.8 %</td>
<td>9.0%</td>
<td>2.4%</td>
</tr>
</tbody>
</table>

- Between-run precision (reproducibility):
In order to evaluate inter-assay reproducibility, one negative and three positive samples (low, medium, high positive) were tested in duplicate in two runs per day over a 20 days period. The ratio (Sample OD / CO) was determined for each sample. Mean Ratio, Standard Deviation (SD) and Coefficient of Variation (%CV) for each of the four specimens are listed in table below:

<table>
<thead>
<tr>
<th>N=80</th>
<th>Negative Sample</th>
<th>Low positive sample</th>
<th>Medium positive sample</th>
<th>High positive sample</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ratio (Sample OD / Cut-Off value)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>0.40</td>
<td>1.63</td>
<td>3.48</td>
<td>6.68</td>
</tr>
<tr>
<td>SD</td>
<td>0.09</td>
<td>0.12</td>
<td>0.22</td>
<td>0.43</td>
</tr>
<tr>
<td>% CV</td>
<td>21.9</td>
<td>7.6</td>
<td>6.3</td>
<td>6.4</td>
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</table>
9.6 CROSS REACTIVITY

129 samples with characteristics which could potentially result in non specific reactions were tested with the Platelia™ HSV 1+2 IgM assay. Results are presented in the following table:

<table>
<thead>
<tr>
<th></th>
<th>Number of samples</th>
<th>Number of Platelia™ HSV 1+2 IgM positive sera</th>
</tr>
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<tbody>
<tr>
<td>HIV</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>CMV IgM</td>
<td>9</td>
<td>0</td>
</tr>
<tr>
<td>Rub IgM</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>EBV IgM</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>Toxo IgM</td>
<td>9</td>
<td>1</td>
</tr>
<tr>
<td>Chlamydia trachomatis</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>VZV IgM</td>
<td>19</td>
<td>4</td>
</tr>
<tr>
<td>Measles IgM</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>Anti-nuclear antibodies (ANA)</td>
<td>10</td>
<td>0</td>
</tr>
<tr>
<td>Mumps IgM</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>Rhumatoid Factors</td>
<td>10</td>
<td>2</td>
</tr>
<tr>
<td>HHV6</td>
<td>9</td>
<td>0</td>
</tr>
<tr>
<td>HPV</td>
<td>3</td>
<td>2</td>
</tr>
</tbody>
</table>

**Note**: equivocal results were not considered in the cross reaction evaluation.

All the positive results with the Platelia™ HSV 1+2 IgM assay were negative with the commercialized IgM EIA assay.

All the sera found positive with Platelia™ HSV 1+2 IgM assay were positive in HSV 1 and/or 2 IgG serology indicating an HSV infectious status except for 2 VZV samples.

10. LIMITATIONS OF THE PROCEDURE

Diagnosis of HSV infection can only be established on the basis of a combination of clinical and biological data. The result of a single test of titration of anti-HSV IgM antibodies does not constitute sufficient proof for the diagnosis of a recent infection by Herpes simplex virus since HSV IgM can also be present in recurrent HSV infections (2).

Considering the antigenic homology of the virus of herpes family, potential cross reactions with other members of this family should not be excluded.
11. QUALITY CONTROL OF THE MANUFACTURER

All manufactured reagents are prepared according to our Quality System, starting from reception of raw material to commercialization of the final product. Each lot is submitted to quality control assessments and is released to the market only after conforming to pre-defined acceptance criteria. The records related to production and controls of each single lot are kept within Bio-Rad.

12. REFERENCES


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<td>The other languages which are required in conformity to the European Directive can be obtained from your local Bio-Rad agent.</td>
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<td>Los otros idiomas que se requieren para la conformidad de la Directiva Europea puede ser obtenida en su oficina local Bio-Rad.</td>
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<td>D</td>
<td>Die anderen Sprachen, die in Übereinstimmung mit der europäischen IVD Direktive benötigt werden, erhalten Sie über Ihre lokale Bio-Rad Niederlassung.</td>
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<td>As restantes línguas, obrigatórias em conformidade com a Directiva Europeia, podem ser obtidas através da subsidiária Bio-Rad mais próxima de si.</td>
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<td>Övriga språk som krävs i enlighet med EG-direktivet kan erhållas från din lokala Bio-Rad-representant.</td>
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<td>De øvrige sprog som kræves i henhold til EU direktiv kan fås ved henvendelse til den lokale Bio-Rad leverandør.</td>
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<td>Οι υπολογισμοί για την κατανόηση της ευρωπαϊκής οδηγίας μπορείτε να τους προμηθευτείτε από τον τοπικό σας αντιπρόσωπο Bio-Rad.</td>
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<td>Tłumaczenie w innych językach które są wymagane w Dyrektywie Unijnej może być otrzymane od lokalnego przedstawiciela firmy Bio-Rad.</td>
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<td>Vertimus, reikalins pagal Europos sejungos direktyvos reikalavimus, jį kitas kalbos galite gauti iš vietinio Bio-Rad atstovo.</td>
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<td>Ostatné jazykové verzie, které sú vyžadované v zhobe s Európskou direktívou, možno obdržať od vlastního lokálu zástupcu Bio-Rad.</td>
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<td>Další jazykové verze vyžadované ve shodě s evropskou direktivou jsou k dispozici u lokálního zástupce firmy Bio-Rad.</td>
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