Product Information

Mouse/Rat HSV-1 IgG ELISA

Catalog Number SE120085
Storage Temperature 2–8 °C

TECHNICAL BULLETIN

Product Description

HSV-1 and 2 are virtually identical, sharing ~50% of their DNA and have over 80% of common antigens. Both types infect the body’s mucosal surfaces, usually the mouth or genitals, and then establish latency in the nervous system. Several recent studies have shown the association of more than a dozen herpes viruses with cancer in man and various animals, for example with lymphoma and with squamous cell carcinoma of the lip and cancer of the cervix. HSV type 1 is the cause of most orofacial herpes and HSV encephalitis; type 2 is the primary cause of initial and recurrent genital herpes and neonatal HSV. Reactivation of latent HSV infection is a frequent complication of immunosuppression due to cancer, transplantation, and AIDS. Asymptomatic genital shedding of HSV-2 is more common than HSV-1 and occurs more frequently during the first 3 months after acquisition of primary type 2 disease than during later periods. The presence of HSV IgG antibody is indicative of previous exposure. A significant increase in HSV IgG is an indicative of reactivation, current or recent infection. IgM antibody is present after primary HSV infection.

The effect of virus dose and animal age on the appearance of acute and latent neurologic infection by HSV-1 and HSV-2 was studied in Balb/c and ICR mice inoculated in the footpad. At low viral doses, HSV-2 was found to be 1,500 times more neurovirulent than HSV-1. The Mp strain of herpes simplex virus type 1 (HSV1) induced a persistent infection in the mouse C 1300 neuronal cell line (clone N 115). C 1300 cultures infected at an MOI of 0.01 or 0.001 survived the initial infection and continued to produce infectious virus and viral antigens for 185 days and 31 days, respectively.

The Mouse/Rat HSV-1 IgG ELISA kit is an enzyme linked immunosorbent Assay (ELISA) used for the detection of IgG class antibodies to HSV-1 in mouse/rat. Diluted serum is added to wells coated with purified antigen. IgG specific antibody, if present, binds to the antigen. All unbound materials are washed away and the enzyme conjugate is added to bind to the antibody-antigen complex, if present. Excess Enzyme Conjugate is washed off and substrate is added. The plate is incubated to allow the oxidation of the substrate by the enzyme. The intensity of the color generated is proportional to the amount of IgG specific antibody in the sample.

Components

<table>
<thead>
<tr>
<th>Materials provided</th>
<th>96 tests</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microwell coated with HSV-1 antigen</td>
<td>12 x 8 x 1</td>
</tr>
<tr>
<td>Sample Diluent: 1 bottle (ready to use)</td>
<td>22 mL</td>
</tr>
<tr>
<td>Calibrator: 1 vial (ready to use)</td>
<td>1 mL</td>
</tr>
<tr>
<td>Positive Control: 1 vial (ready to use)</td>
<td>1 mL</td>
</tr>
<tr>
<td>Negative Control: 1 vial (ready to use)</td>
<td>1 mL</td>
</tr>
<tr>
<td>Enzyme conjugate: 1 bottle (ready to use)</td>
<td>12 mL</td>
</tr>
<tr>
<td>TMB Substrate: 1 bottle (ready to use)</td>
<td>12 mL</td>
</tr>
<tr>
<td>Stop Solution: 1 bottle (ready to use)</td>
<td>12 mL</td>
</tr>
<tr>
<td>Wash concentrate 20x: 1 bottle</td>
<td>25 mL</td>
</tr>
</tbody>
</table>

Reagents and Equipment Required but Not Provided.
1. Distilled or deionized water
2. Precision pipettes
3. Disposable pipette tips
4. ELISA reader capable of reading absorbance at 450 nm
5. Absorbent paper or paper towel
6. Graph paper
Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions
Sample Preparation
Collect blood specimens and separate the serum. Specimens may be refrigerated at 2–8 °C for up to seven days or frozen for up to six months. Avoid repetitive freezing and thawing.

20x Wash Buffer Concentrate
Prepare 1x Wash buffer by adding the contents of the bottle (25 mL, 20x) to 475 mL of distilled or deionized water. Store at room temperature (18–26 °C).

Storage/Stability
Store the kit at 2–8 °C.

Procedure
Notes: The components in this kit are intended for use as an integral unit. The components of different lots should not be mixed.

Optimal results will be obtained by strict adherence to the test protocol. Precise pipetting as well as following the exact time and temperature requirements is essential.

The test run may be considered valid provided the following criteria are met:

1. The O.D. of the Calibrator should be >0.250.
2. The Ab index for Negative control should be <0.9.
3. The Ab Index for Positive control should be >1.2.

Bring all specimens and kit reagents to room temperature (18–26 °C) and gently mix.

1. Place the desired number of coated strips into the holder.
2. Negative control, positive control, and calibrator are ready to use. Prepare 21-fold dilution of test samples by adding 10 µL of the sample to 200 µL of Sample Diluent. Mix well.
3. Dispense 100 µL of diluted sera, calibrator, and controls into the appropriate wells. For the reagent blank, dispense 100 µL of Sample Diluent in 1A well position. Tap the holder to remove air bubbles from the liquid and mix well. Incubate for 20 minutes at room temperature.
4. Remove liquid from all wells. Wash wells three times with 300 µL of 1x Wash buffer. Blot on absorbent paper or paper towel.
5. Dispense 100 µL of Enzyme Conjugate to each well and incubate for 20 minutes at room temperature.
6. Remove Enzyme Conjugate from all wells. Wash wells three times with 300 µL of 1x Wash buffer. Blot on absorbent paper or paper towel.
7. Dispense 100 µL of TMB substrate and incubate for 10 minutes at room temperature.
8. Add 100 µL of Stop Solution.
9. Read O.D. at 450 nm using ELISA reader within 15 minutes. A dual wavelength is recommended with reference filter to 600–650 nm.
Results

Calculations
1. Check Calibrator Factor (CF) value on the calibrator bottle. This value might vary from lot to lot. Make sure the value is checked on every kit.
2. Calculate the cut-off value: Calibrator OD x Calibrator Factor (CF).
3. Calculate the Ab (Antibody) Index of each determination by dividing the O.D. value of each sample by cut-off value.

Example of typical results:
Calibrator mean OD = 0.8  
Calibrator Factor (CF) = 0.5  
Cut-off Value = 0.8 x 0.5 = 0.400  
Positive control O.D. = 1.2  
Ab Index = 1.2/0.4 = 3  
Patient sample O.D. = 1.6  
Ab Index = 1.6/0.4 = 4.0

Note: The test results obtained using this kit cannot discriminate between HSV-1 and HSV-2 infection due to high cross reactivity between the two viruses.

Lipemic or hemolyzed samples may cause erroneous results.

Interpretation
The following is intended as a guide to interpretation of HSV-1 IgG antibody index (Ab Index) test results; each laboratory is encouraged to establish its own criteria for test interpretation based on sample populations encountered.

<0.9 – No detectable antibody to HSV-1 IgG by ELISA  
0.9–1.1 – Borderline positive. Follow-up testing is recommend if clinically indicated.  
>1.1 – Detectable antibody to HSV-1 IgG by ELISA

Converting of Ab Index to IU/mL
As an option, Ab index may be converted to IU/mL by multiplying Ab index by 100. IU/mL values may then be interpreted as follows:

<90 IU/mL – No detectable antibody to HSV-1 IgG by ELISA  
90–110 IU/mL – Borderline positive. Follow-up testing is recommend if clinically indicated.  
>110 IU/mL – Detectable antibody to HSV-1 IgG by ELISA

Product Profile

Precision

Intra Assay Study

<table>
<thead>
<tr>
<th>Serum</th>
<th>No. of Replicates</th>
<th>Mean</th>
<th>Standard Deviation</th>
<th>Coefficient of Variation %</th>
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</thead>
<tbody>
<tr>
<td>1</td>
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<td>1.23</td>
<td>0.06</td>
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<td>16</td>
<td>0.33</td>
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<td>6.06</td>
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Inter Assay Study

<table>
<thead>
<tr>
<th>Serum</th>
<th>No. of Replicates</th>
<th>Mean</th>
<th>Standard Deviation</th>
<th>Coefficient of Variation %</th>
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<tbody>
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<td>1</td>
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<td>1.77</td>
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<tr>
<td>2</td>
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<td>9.47</td>
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<tr>
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<td>10</td>
<td>0.21</td>
<td>0.02</td>
<td>14.2</td>
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References

Al,CH,MAM 10/14-1