

Antibody Datasheet

Product name

Herpes Simplex Virus Type 1 + 2 Glycoprotein D antibody (3-1D)

Product description

Complement-independent neutralizing monoclonal antibody to HSV-gD (see next page for details)

Catalog Number

EVHM0401-100

Source

Human (recombinant production in CHO-K1)

Clonality and Clone name

Monoclonal, 3-1D

Isotype

IgG1 Kappa

Form Supplied and Size

Liquid, 100 µg

Concentration and storage buffer

1 mg/mL in Phosphate buffer saline pH7.4 (containing no preservative)

Storage

Antibody can be kept at 4°C for up to 1 month and should be kept at -20°C or below for long-term storage. To avoid repeated freeze thaw cycles, antibody should be aliquoted before frozen.

Purification

Purified by protein A chromatography. The purity is greater than 95% by SDS-PAGE.

Antigen for Screening

Recombinant HSV1 (F strain) glycoprotein D-expressing CHO-K1 and recombinant HSV2 (G strain) glycoprotein D-expressing CHO-K1

Epitope

Epitope has not been determined.

Applications

ICC/IF, Neutralization assay. Other applications have not been tested.

Limitations

This product is to be used for research purposes only.

Background information

Herpes Simplex Virus (HSV) is a ubiquitous human alpha-herpesvirus that establishes life-long latent infections in human following the primary infection. There are two types of HSV, HSV-1 and HSV-2. The sero-prevalence of HSV-1 is more common than HSV-2. HSV-1 infection commonly develops herpes labialis (cold sore or fever blisters) and can also cause genital herpes. HSV-2 generally infects genital area and can cause genital herpes. It has been shown that four of HSV-encoded glycoproteins are necessary and sufficient to facilitate membrane fusion between viral and host membranes. HSV glycoprotein D (gD) binds to cellular receptors that are responsible for viral penetration and is essential for viral entry into host cell.

Immunogen and Recombinant Production Host

This antibody was generated from a healthy individual by a method based on the Epstein-Barr virus transformation of peripheral blood mononuclear cells followed by the isolation of antibody-producing cells. The antibody reactivity for the target antigen was screened by immunofluorescent staining by using CHO-K1 cells displaying recombinant HSV gD. The antibody genes were cloned from the antibody-producing cells and introduced into CHO-K1 cells for antibody production.

Application Note

Recommended Starting Dilutions:

For ICC/IF: Use at 1:500 – 1:4000

Not yet tested in other applications.

The optimal working dilution should be determined experimentally by the end user.

Neutralization assay

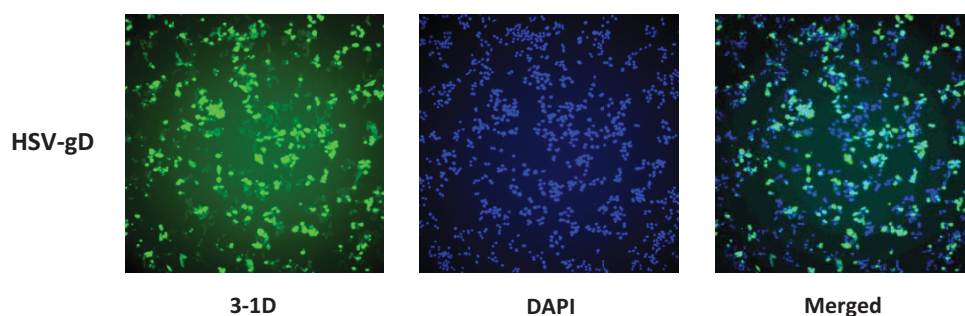
The 50% and 90% of inhibitory concentration (IC_{50} and IC_{90}) values of the antibody against HSV-1 were ≥ 284 ng/mL and ≥ 809 ng/mL, and those against HSV-2 were ≥ 723 ng/mL and ≥ 2469 ng/mL, respectively.

The IC_{50} and IC_{90} were determined as described below.

1. Plate Vero cells in a tissue plate with growth medium, and incubate them at 37 °C, 5% CO₂.
2. Check the plate to confirm 100% confluency and even cell distribution.
3. Prepare serial dilution of antibodies, and mix with an equal volume of virus diluent. Include the following controls: no antibody control, normal IgG control and no virus control in defined wells.
4. Gently agitate the virus-antibody mixtures, and incubate for 1 hour at 37 °C, 5% CO₂.
5. Remove culture media and transfer co-incubated antibody-virus mixture to wells containing monolayers of the target cells.
6. Incubate for 1 hour at 37 °C, 5% CO₂.
7. Remove culture medium from each well and wash each well twice with assay medium.
8. Add assay medium to each well.
9. Incubate the plate under appropriate condition.
10. Remove assay medium from the well.
11. Fix cells and detect virus-infected cells by immunofluorescent staining for the determination of IC_{50} and IC_{90} .

Consult the available literature for the best system for your intended assay.

Immunofluorescence Results



Immunofluorescent staining was performed by using HSV-gD-expressing CHO-K1. Brief protocol is described below.

- 1) Transfect plasmid vector that contains HSV-gD gene into CHO-K1.
- 2) Twenty four hours post-transfection, fix cells with 4% paraformaldehyde-PB.
- 3) Permeabilized cells with 0.2% Tween-PBS for a few minutes.
- 4) Dilute 6-10E antibody and use as primary antibody for 1hr.
- 5) Use anti-human IgG (gamma chain)-FITC as secondary antibody for 1hr.
- 6) Dilute DAPI and treat for 5 min.
- 7) Wash with PBS four times.
- 8) Observe images by fluorescence microscopy.



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