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SARS-CoV-2 (A222V, D614G) Pseudoviral Particles

CATALOG NUMBER: SCV2-PsV-222V, 2x 5 mL

Description

It has been known that SARS coronavirus 2 (SARS-CoV-2) uses human ACE2 as the entry receptor and human proteases as the entry activators. The virus surface spike (S) protein mediates SARS-CoV-2 entry into cells. To fulfill its function, SARS-CoV-2 spike binds to the human ACE2 (hACE2) receptor through its receptor-binding domain (RBD) and then proteolytically activated by host proteases.

Our SARS-CoV-2 Pseudoviral Particles are replication-deficient MLV pseudotyped with the SARS-CoV-2 spike protein. They also contain the ORF for firefly luciferase as a reporter. They establish a pseudovirus entry assay for SARS-CoV-2 as the S protein mediated cell entry can be conveniently measured via the luciferase reporter activity. This pseudovirus assay isolates the SARS-CoV-2 viral entry from other steps of the viral infection cycle.

A new SARS-CoV-2 strain with an amino acid change at position 222 from Ala to Val besides the change at position 614 from Asp to Gly in the viral S protein predominated over time in locales where it was found. To support the study of this new SARS-CoV-2 strain, we established this **SARS-CoV-2-614G-A222V Pseudoviral Particles** with the spike protein carrying the 222V and 614G genotype (Genbank Accession #QHD43416.1, p.222 A->V, p.614 D->G)

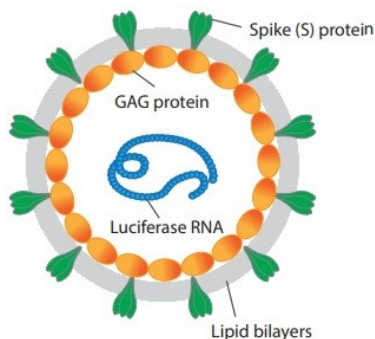


Figure 1. Illustration of the replication-deficient MLV particle pseudotyped with SARS-CoV-2 Spike protein

Applications

Coupled with firefly luciferase assay kit (Catalog # [CA-L165](#)), the pseudovirus infection generates robust chemiluminescent signals, 1) for screening potential inhibitor to block SARS-CoV-2 entry and viral protein translation; 2) for measuring the activity of and screening for neutralizing antibody against SARS-CoV-2-614G-A222V (refer to [the Neutralization Assay Application Note](#)).

Features

- **Robust:** Excellent signal to noise (basal) ratio
- **Easy to use:** Amenable to HTS format (96-well, 384-well and 1536-well format)

Contents

10 ml (2 tubes, 5 mL/tube), for 2 multi-well plates

Storage

Store at -70°C

Shelf Life:

Six months from the date of shipping when store at -70 °C



ASSAY PROTOCOL

Note: requires a luciferase assay reagent (Catalog # [CA-L165](#)).

Cell Infection:

1. Count HEK293-ACE2 cells (Catalog # [CL-hACE2-001](#)) to be infected and seed ~20K cells per well into 96-well plates (50 μ l per well) DMEM with 10% HyClone™ FetalClone™ II Serum (no antibiotics) or 5K cells per well into 384-well plates (15 μ l per well).
2. Culture cells overnight to make sure the cells stably adhere to the plates.
3. On the 2nd day, remove media, add 50 μ l SARS-CoV-2 pseudoviral particles into each well (12.5 μ l for 384-well plate). Spin at 700 rpm for 15 min at 4°C.

Note: thaw the pseudoviral particles immediately before use (take about 1 hour to thaw in ice) and use it within 2 hours.

4. Incubate for 2 hrs at 37 °C.
5. Add 50 μ l DMEM with 10% FC into each well (12.5 μ l for 384-well plates).
6. Incubate for 48 hrs at 37 °C.

Measurement of Luciferase Activity in Infected cells

1. Remove supernatant.
2. Add 100 μ l eEnzyme's luciferase assay reagent (20 μ l for 384-well plates).
3. Read in a luminescence plate reader, and record the data.

Data Analysis

