SARS-CoV-2 Pseudoviral Particles, Gamma Variant (Brazil P.1)

CATALOG NUMBER: SCV2-PsV-BR, 5 mL

Description

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

Our **SARS-CoV-2-BR Pseudoviral Particles** are replication-deficient MLV pseudotyped with the SARS-CoV-2 spike protein of the Gamma variant (also known as Brazil P.1, VOC-202101/02, 20J/501Y.V3) (GISAID sequence accession # EPI_ISL_833172). They also contain the ORF for firefly luciferase as a reporter. They establish a pseudovirus cell entry assay mediated by the SARS-CoV-2 spike protein that can be conveniently measured via luciferase reporter activity. This pseudovirus assay isolates the SARS-CoV-2 viral entry from other steps of the viral infection cycle.

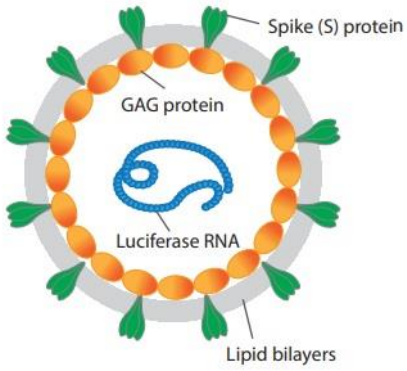

Figure 1. Illustration of the replication-deficient MLV particle pseudotyped with SARS-CoV-2 Spike protein

Reference:

Genomic characterisation of an emergent SARS-CoV-2 lineage in Manaus: preliminary findings. *https://virological.org/t/genomic-characterisation-of-an-emergent-sars-cov-2-lineage-in-manaus-preliminary-findings/586*

Applications

Our Pseudovirus Particles generate robust chemiluminescent signals in cell assays when coupled with our firefly luciferase assay kit (Catalog # CA-L165), useful for 1) screening potential inhibitor to block SARS-CoV-2 entry and viral protein translation; 2) measuring the activity of and screening for neutralizing antibody against SARS-CoV-2-BR (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

5 ml, for one multi-well plate; PP per mL > 1.0E+07

Storage

Upon receiving this item, store at -70 °C right away. Thaw* before immediate use.
Note: read the instruction for thawing in the following protocol carefully. Do not aliquot and refreeze.

Shelf Life:

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

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ASSAY PROTOCOL

Note: require a luciferase assay reagent (Catalog # CA-L165).

Cell Infection:

1. Count HEK293-ACE2 cells (Catalog # CL-hACE2-002) to be infected and seed ~20K cells per well into 96-well plates (50 µl per well) DMEM with 10% HyClone™ FetalClone™ II Serum (no antibiotics) or 5K cells per well into 384-well plates (15 µl 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 µl SARS-CoV-2-BR Pseudoviral Particles* into each well (12.5 µl for 384-well plate). Spin at 700 rpm for 15 min at 4°C.

   Note: thaw the pseudoviral particles quickly in the room temperature water (< 30 minutes, do not shake) and use right away. Discard the unused portion (do not re-freeze or leave it on ice for later use).
4. Incubate for 2 hrs at 37 °C.
5. Add 50 µl DMEM with 10% FC into each well (12.5 µl for 384-well plates).
6. Incubate for 42 hrs at 37 °C.

Measurement of Luciferase Activity in Infected Cells

1. Do not remove medium. Add 100 µl eEnzyme's luciferase assay WORKING SOLUTION (25 µl for 384-well plate) directly into each well. Refer to the protocol of "Firefly Luciferase Assay Kit" (eEnzyme Cat.# CA-L165).
2. Read in a luminescence plate reader and record the data.

Data Analysis

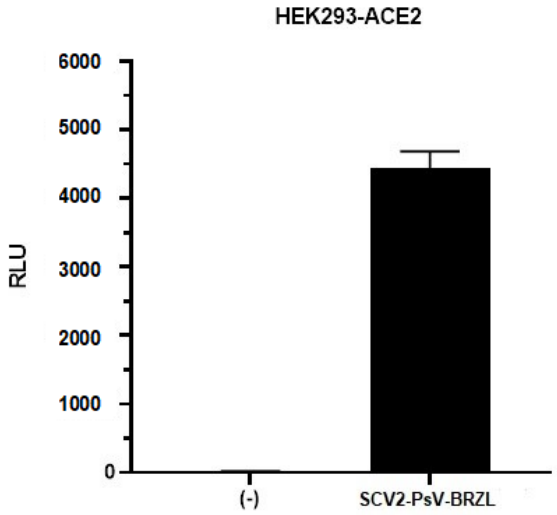

Figure 2. Pseudoviral Particle (PP) Infection Assays
SARS-CoV-2-BR variant pseudoviral particles on HEK293-ACE2 cells in 384-well format
(Luminescence plate reader: PerkinElmer WALLAC VICTOR^2 1420 Multilabel Counter)
Legends: SCV2-PsV-BRZL: SARS-CoV-2 Brazil variant MLV pseudovirus particles (SCV2-PsV-BR)
(-): MLV control (w/o envelope spike protein) (Catalog # PsV-001)