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Zuschläge

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- Trockeneiszuschlag
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SARS-CoV-2 Pseudoviral Particles, 614G

CATALOG NUMBER: SCV2-PsV-614G, 10 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 receptor human ACE2 (hACE2) through its receptor-binding domain (RBD) and is proteolytically activated by human 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 614 from Asp to Gly in the viral S protein predominated over time in locales where it was found. It is reported by the Choe lab in The Scripps Research Institute that this currently dominate new strain with the D614G variation in the S protein “reduces S1 shedding and increases infectivity”. To support the study of this new SARS-CoV-2 strain, we established the SARS-CoV-2 Pseudoviral Particles with the new S variant (614G).

Applications

Work perfectly for Luc pseudovirus to get robust signal, 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.

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

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ASSAY PROTOCOL

Note: requires a luciferase assay reagent (any commercially available)

Cell Infection:

1. Count Vero E6 cells/HEK293-ACE2 cells to be infected and seed ~20K cells per well into 96-well plates (50 µl per well) DMEM with 10% FC (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 pseudoviral particles into each well (12.5 µl for 384-well plate). Spin at 700 rpm for 15 min at 4°C.
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 48 hrs at 37 °C.

Measurement of Luciferase Activity in Infected cells

1. Remove supernatant.
2. Add 100 µl Luciferase assay reagent (20 µl for 384-well plates).
3. Read in a luminescence plate reader, and record the data.

Data Analysis

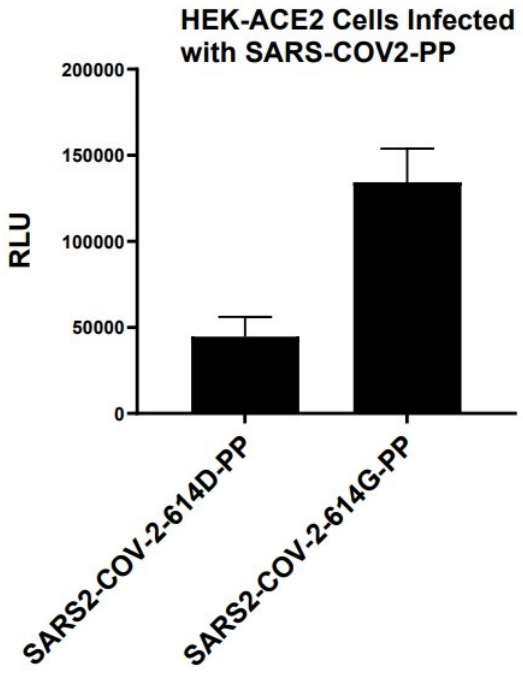

Figure 1. Pseudoviral Particle (PP) Infection Assays

SARS2-CoV-2-614D-pp denotes the PP pseudotyped with the 614D (original) spike protein (Cat.# SCV2-PsV-001).
SARS2-CoV-2-614G-pp denotes the PP pseudotyped with the 614G spike protein (Cat.# SCV2-PsV-614G).

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