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Produktinformation

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- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

linkedin.com/company/szaboscandic

Description

The pandemic coronavirus disease 2019 (COVID-19) is caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). As the first step of the viral replication, the virus attaches to the host cell surface before entering the cell. The viral Spike protein recognizes and attaches to the Angiotensin-Converting Enzyme 2 (ACE2) receptor found on the surface of type I and II pneumocytes, endothelial cells, and ciliated bronchial epithelial cells. Drugs targeting the interaction between the Spike protein and ACE2 may offer protection against the viral infection.

A variant called B.1.351 was first identified in the fall of 2020 in the Republic of South Africa. This South African variant, also known as 501Y.V2, has many mutations that may lead to higher transmissibility and infectivity. The Spike (B.1.351 Variant) (SARS-CoV-2) Pseudotyped Lentivirus were produced with SARS-CoV-2 B.1.351 Variant Spike (Genbank Accession #QHD43416.1 with B.1.351 mutations; see below for details) as the envelope glycoproteins instead of the commonly used VSV-G. These pseudovirions contain the enhanced green fluorescent protein (eGFP) gene driven by a CMV promoter (Figure 1), therefore, the spike-mediated cell entry can be determined via eGFP fluorescence. The Spike (B.1.351 Variant) (SARS-CoV-2) Pseudotyped Lentivirus can be used to measure the activity of neutralizing antibody against SARS-CoV-2 (B.1.351) variant in a Biosafety Level 2 facility.

Spike Mutations in the B.1.351 Variant

L18F
D80A
D215G
R246I
K417N
E484K
N501Y
D614G
A701V

Application

1. Study the mechanism of viral transduction of SARS-CoV-2 (B.1.351 variant)
2. Screening for neutralizing antibodies for SARS-CoV-2 Spike (B.1.351 variant) and ACE2.

Formulation

The lentiviruses were produced from HEK293T cells. Supplied in medium containing 90% DMEM + 10% FBS.

Titer

The titer will vary with each lot; the exact value is provided with each shipment.

Storage

Lentiviruses are shipped with dry ice. For long term storage, it is recommended to store the virus at -80°C. Avoid repeated freeze-thaw cycles. Titers can drop significantly with each freeze-thaw cycle.

Biosafety

None of the HIV genes (gag, pol, rev) will be expressed in the transduced cells, as they are expressed from packaging plasmids lacking the packing signal. Although the pseudotyped lentiviruses are replication-incompetent, they require the use of a Biosafety Level 2 facility. BPS recommends following all local federal, state, and institutional regulations and using all appropriate safety precautions.

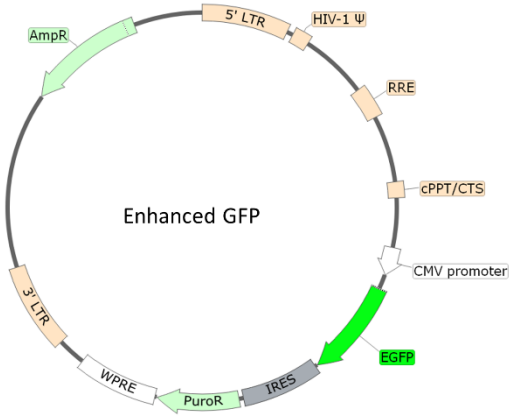

Figure 1. Schematic of the eGFP Reporter in Spike (B.1.351 Variant) (SARS-CoV-2) Pseudotyped Lentivirus

Materials Required but Not Supplied

These materials are not supplied with this lentivirus but are necessary to follow the designed protocol. BPS Bioscience media and reagents are all validated and optimized for use with this lentivirus and are highly recommended for best results.

Name	Ordering Information
Thaw Medium 1	BPS Bioscience, #60187
ACE2- HEK293 Recombinant Cell Line	BPS Bioscience, #79951
96-well white clear-bottom assay plate	Corning, #3610

Assay Protocol

The following protocol is a general guideline for transducing ACE2-HEK293 cells using Spike (B.1.351 Variant) (SARS-CoV-2) pseudotyped lentivirus (eGFP reporter). The optimal transduction conditions (e.g. MOI, concentration of polybrene, time of assay development) should be optimized according to the cell

type and the assay requirements. In most cell types, the expression of the reporter gene can be measured approximately 48-72 hours after transduction.

1. Day 1: Harvest ACE2-HEK293 cells from culture and seed cells at a density of 5,000-10,000 cells per well into white clear-bottom 96-well microplate in 50 µl of Thaw Medium 1 (BPS Bioscience, #60187). Incubate cells at 37°C with 5% CO_2 overnight.

To demonstrate transduction is dependent on ACE2, the same number of HEK293 parental cells can be seeded in Thaw Medium 1 as control cells.

2. Day 2: Add 20 µl of Spike (B.1.351 Variant) (SARS-CoV-2) Pseudotyped Lentivirus (eGFP reporter) into each well.

Optional: Add polybrene to each well at a final concentration of 5 µg/ml.

Incubate the plates at 37°C with 5% CO_2 overnight.

Alternatively, seeding cells and the transduction can be performed on the same day.

3. Day 3: Remove the medium containing the lentivirus from the wells. Add 50 µl of fresh Thaw Medium 1 to each well.

If neither the polybrene nor the lentivirus adversely affects the target cells, it is not necessary to change the medium on Day 3. The target cells can be incubated with the virus for 48-72 hours before changing medium.

4. Day 4-5, approximately 48-72 hours after transduction, the expression of eGFP in the target cells was examined under a fluorescence microscopy.

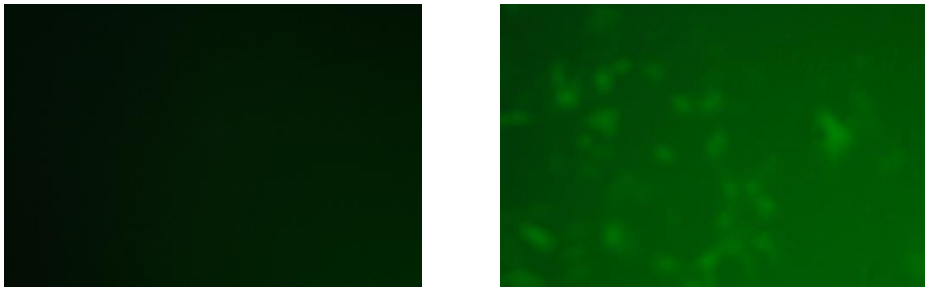

Figure 2. Transduction of ACE2-HEK293 cells using Spike (SARS-CoV-2, B.1.351 variant) pseudotyped lentivirus (eGFP reporter). Approximately 8,000 cells/well of ACE2-HEK293 cells (right) or HEK293 parental cells (left) were seeded and transduced on the same day with 20 µl/well of Spike (SARS-CoV-2, B.1.351 variant) pseudotyped lentivirus (eGFP reporter). After 66 hours of transduction, the expression of eGFP in the target cells was observed under a fluorescence microscope.

License Disclosure
Visit bpsbioscience.com/license for the label license and other key information about this product.

Troubleshooting Guide
Visit bpsbioscience.com/lentivirus-faq for detailed troubleshooting instructions. For all further questions, please email support@bpsbioscience.com.

Related Products

Spike (P.1 Variant) (SARS-CoV-2) Pseudotyped Lentivirus (eGFP Reporter)	78159	500 µl x2
Spike (B.1.1.7 Variant) (SARS-CoV-2)Pseudotyped Lentivirus (eGFP Reporter)	78158	500 µl x2
Spike Pseudotyped Lentivirus (eGFP Reporter)	79981	500 µl x2
Bald Lentiviral Pseudoviron (eGFP Reporter)	79987	500 µl x2
Spike Pseudotyped Lentivirus (Luciferase-eGFP Dual Reporter)	79982	500 µl x2
Bald Lentiviral Pseudoviron (Luciferase-eGFP Dual Reporter)	79988	500 µl x2
Spike Pseudotyped Lentivirus (Luciferase Reporter)	79942	500 µl x2
Bald Lentiviral Pseudovirion (Luciferase Reporter)	79943	500 µl x2
Spike (B.1.1.7 Variant) (SARS-CoV-2) Pseudotyped lentivirus (Luc reporter)	78112	500 µl x2
Spike (P.1 Variant) (SARS-CoV-2) Pseudotyped lentivirus (Luc reporter)	78144	500 µl x2
Spike (B.1.351 variant) (SARS-CoV-2) Pseudotyped lentivirus (Luc reporter)	78142	500 µl x2
ACE2-HEK293 Recombinant Cell Line	79951	2 vials
Thaw Medium 1	60187	100 ml