Description

Severe acute respiratory syndrome (SARS) was the first new infectious disease identified in the twenty-first century. It is a viral respiratory disease caused by severe acute respiratory syndrome coronavirus (SARS-CoV-1). The first known cases occurred in November 2002, and these viral infections led to the 2002–2004 SARS outbreak. Since 2004, no cases of SARS-CoV-1 have been reported worldwide. A virus very similar to SARS-CoV-1 was discovered in late 2019. This virus, named severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), is the causative pathogen of COVID-19, the spread of which started the COVID-19 pandemic.

SARS-CoV-1 attaches to the host cell surface before entering the cell. The Spike protein on the virus recognizes and binds to the Angiotensin-Converting Enzyme 2 (ACE2) receptor found on the surface of human airway epithelia as well as lung parenchyma. Drugs targeting the interaction between the Spike protein of SARS-CoV-1 and ACE2 may offer protection against the viral infection.

The Spike (SARS-CoV-1) Pseudotyped Lentiviruses were produced with SARS-CoV-1 Spike (Genbank Accession #YP_009825051.1) as the envelope glycoproteins instead of the commonly used VSV-G. These pseudovirions contain the eGFP gene driven by a CMV promoter (Figure 1), therefore, the spike-mediated cell entry can be determined via eGFP fluorescence. The Spike (SARS-CoV-1) pseudotyped lentivirus can be used to measure the activity of a neutralizing antibody against SARS-CoV-1 in a cellular context, using a Biosafety Level 2 facility.

As shown in Figure 2, the Spike (SARS-CoV-1) pseudovirus has been validated for use with ACE2-HEK293 target cells (which overexpress ACE2; BPS Bioscience #79951).

Applications

Screening for neutralizing antibodies against SARS-CoV-1 Spike protein.

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.

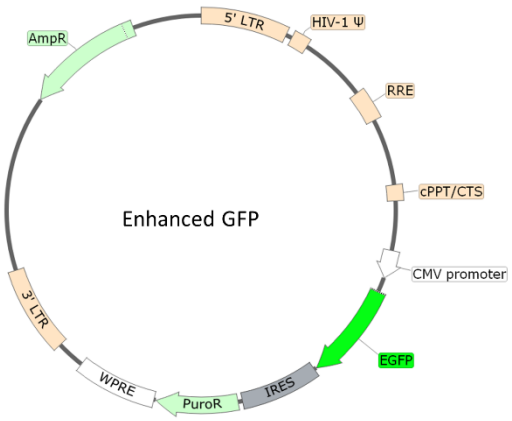

Figure 1. Schematic of the eGFP Reporter in Spike (SARS-CoV-1) Pseudotyped Lentivirus

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 (SARS-CoV-1) 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). Add 1 µl of Spike (SARS-CoV-1) 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.

2. Day 3: Approximately 48-72 hours after transduction, examine the expression of eGFP in the target cells by fluorescence microscopy.

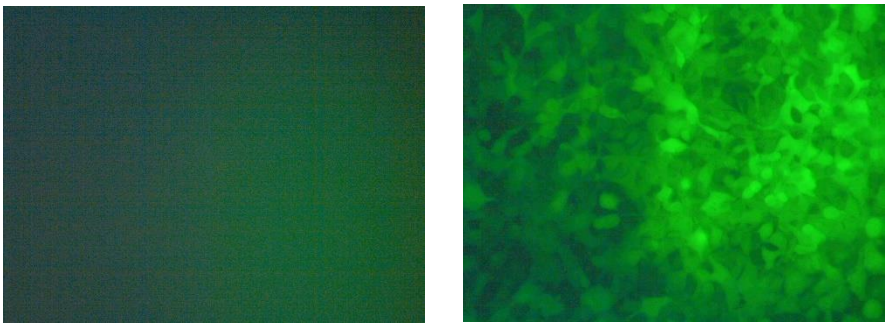

Figure 2. Transduction of ACE2-HEK293 cells using Spike (SARS-CoV-1) Pseudotyped Lentivirus (eGFP Reporter). Approximately 5,000 cells/well of ACE2-HEK293 cells (right) or HEK293 parental cells (left) were seeded and transduced on the same day with 1 µl/well of Spike (SARS-CoV-1) pseudotyped lentivirus (eGFP reporter). After 66 hours of transduction, the expression of eGFP in the target cells was observed under a fluorescence microscope.

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

Related Products

Products	*Catalog #*	*Size*
Bald Lentiviral Pseudovirion (eGFP Reporter)	79987	500 µl x 2
ACE2 - HEK293 Recombinant Cell Line	79951	2 vials
Spike (SARS-CoV-1) Pseudotyped Lentivirus (Luc Reporter)	78614	500 µl x 2
Spike (SARS-CoV-2) Pseudotyped Lentivirus (eGFP Reporter)	79981	500 µl x 2
Spike Variants (SARS-CoV-2) Pseudotyped Lentivirus Pack (Luciferase Reporter)	78616	100 µl x 12
Spike (B.1.1.529 BA.1, Omicron Variant) (SARS-CoV-2) Pseudotyped Lentivirus (eGFP Reporter)	78349	500 µl x 2
Spike (BA.1.1, Omicron Variant R346K) (SARS-CoV-2) Pseudotyped Lentivirus (eGFP Reporter)	78624	500 µl x 2
Spike (BA.2, Omicron Variant) (SARS-CoV-2) Pseudotyped Lentivirus (eGFP Reporter)	78626	500 µl x 2