

Description

The Claudin-4 Lentiviruses are replication incompetent, HIV-based, VSV-G pseudotyped lentiviral particles that are ready to transduce nearly all types of mammalian cells, including primary and non-dividing cells. The particles contain a human Claudin-4 (NM_001305.5) driven by an EF1A promoter and a puromycin selection marker (Figure 1).

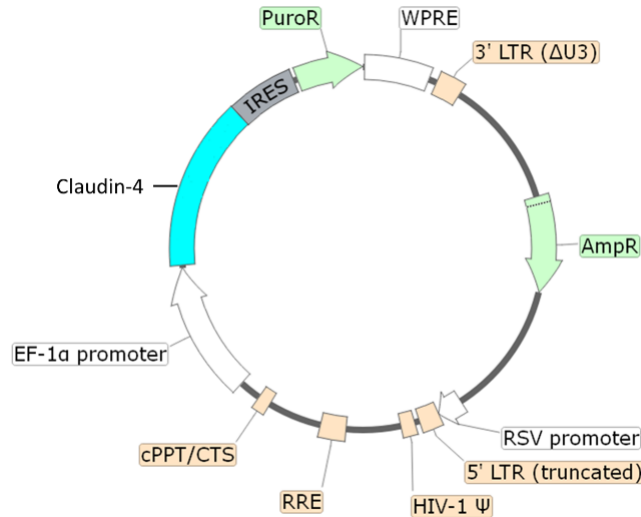


Figure 1: Schematic of the lenti-vector used to generate the Claudin-4 Lentivirus

Background

Claudins are integral membrane proteins and major components of tight junction strands which serve as a physical barrier to prevent solutes and water from passing freely through the paracellular space between epithelial or endothelial cell sheets. Claudin-4 is one of the 27 members of the Claudin family.

Claudin-4 mediates paracellular chloride transport and plays an important renal function through the reabsorption of filtered chloride. The CLDN4 gene is deleted in Williams-Beuren syndrome, leading to multi-systemic neurodevelopmental disorders. The protein acts as a high-affinity receptor for Clostridium perfringens enterotoxin (CPE) during infection by gram-positive Clostridial bacteria, which are responsible for food poisoning and gastrointestinal illnesses. Recent studies indicate that the protein may be overexpressed in ovarian cancer.

Application(s)

Generate stable cell line expressing human Claudin-4 with puromycin selection

Formulation

The lentivirus particles were produced from HEK293T cells. They are supplied in cell culture medium containing 90% DMEM + 10% FBS.

Titer

Two vials (500 μ l x 2) of lentivirus at a titer $\geq 10^7$ TU/ml. 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 lentiviruses at -80°C . Avoid repeated freeze/thaw cycles. Titers can drop significantly with each freeze/thaw cycle.

Biosafety

The lentiviruses are produced with SIN (self-inactivation) lentivector which ensures self-inactivation of the lentiviral construct after transduction and integration into the genomic DNA of the target cells. 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 and are not present in the lentivirus particle. Although the pseudotyped lentiviruses are replication-incompetent, they require the use of a Biosafety Level 2 facility. BPS Bioscience recommends following all local federal, state, and institutional regulations and using all appropriate safety precautions.

Notes

To generate a Claudin-4 stable cell line, remove the growth medium 48 hours after transduction and replace it with fresh growth medium containing the appropriate amount of puromycin (as pre-determined from a killing curve) for antibiotics selection of transduced cells. Visit: <https://bpsbioscience.com/cell-line-faq> for guidelines on performing a kill curve.

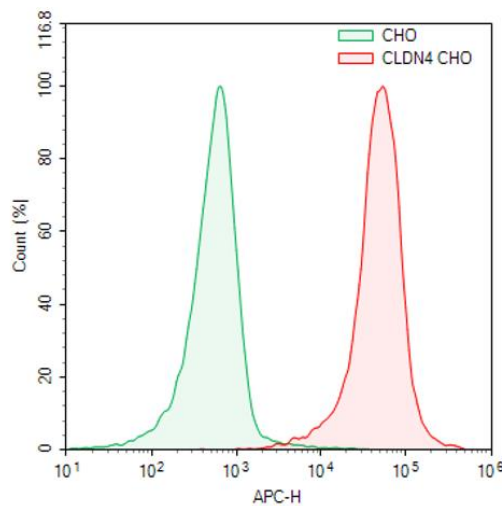
Figures and Validation Data

Figure 2. Transduction of CHO-K1 using Claudin-4 Lentivirus.

Approximately 50,000 CHO-K1 cells were transduced with 500,000 TU of Claudin-4 lentiviruses. After 66 hours of transduction, the cells were selected with 5 µg/ml of puromycin. The puromycin-resistant cell pool was stained using Claudin-4 APC-conjugated Antibody (R&D Systems #FAB4219A) and analyzed by flow cytometry.

Sequence

Human Claudin-4 (NM_001305.5)

MASMGLQVMGIALAVLGWLAVMLCCALPMWRVTAFIGSNIVTSQTIWEGLWMNCVVQSTGQMQCKVYDSSLALPQDLQA
ARALVIISIIVAALGVLLSVVGGKCTNLEDESAKAKTMIVAGVVFLAGLMVIVPVSWTAHNIIQDFYNPLVASGQKREMGASLYV
GWAASGLLLGGLLCCNCPRTDKPYSAKYSAAARSAAASNYV

Troubleshooting Guide

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

Related Products

<i>Products</i>	<i>Catalog #</i>	<i>Size</i>
Trop2 Lentivirus	78710	500 µl x 2
GPC3 Lentivirus	78711	500 µl x 2
Nectin-4 Lentivirus	78712	500 µl x 2
BCMA Lentivirus	78714	500 µl x 2
FcRL5 Lentivirus	78715	500 µl x 2
GPRC5D Lentiviruses	78716	500 µl x 2
Claudin-9 Lentivirus	78721	500 µl x 2
Claudin-3 Lentivirus	78722	500 µl x 2
LYPD1 Lentivirus	78724	500 µl x 2
PSMA Lentivirus	78726	500 µl x 2