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Produktinformation

- Forschungsprodukte & Biochemikalien
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- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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See the following pages for more information!

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

CIITA (class II major histocompatibility complex transactivator) acts as a coactivator for MHC (major histocompatibility complex) class II-specific gene expression and negatively regulates the IL-4 gene promoter during T cell differentiation. IFN-γ (interferon-gamma) induces CIITA gene expression via Janus kinase 1) and Stat1 (Signal transducer and activator of transcription 1) pathways. The GTP-binding and acidic, proline-serine threonine-rich regions appear to be required for CIITA activity. Defects of CIITA has been implicated as causes of bare lymphocyte syndrome (BLS), which is characterized by the absence of MHC class II transcription and severe immunodeficiencies.

The CIITA CRISPR/Cas9 Lentiviruses are replication incompetent, HIV-based, VSV-G pseudotyped lentiviral particles that are ready to infect almost all types of mammalian cells, including primary and non-dividing cells. The particles contain a CRISPR/Cas9 gene driven by an EF1a promoter, along with 5 sgRNA (single guide RNAs) targeting human CIITA driven by a U6 promoter (Figures 1 and Table 1).

Unlike CIITA CRISPR/Cas9 Lentivirus (Integrating) (BPS Bioscience #78435), the CIITA CRISPR/Cas9 Lentivirus (Non-Integrating) is made with a mutated Integrase, resulting in only transient expression of the Cas9 and CIITA targeting sgRNA. This transient expression minimizes potential off-target effects caused by either prolonged expression or random integration of Cas9 and the sgRNA. A short round of puromycin selection right after transduction may increase knockout efficiency, however puromycin should not be used for more than 48 hours post-transduction due to the transient nature of expression using the non-integrating lentivirus.

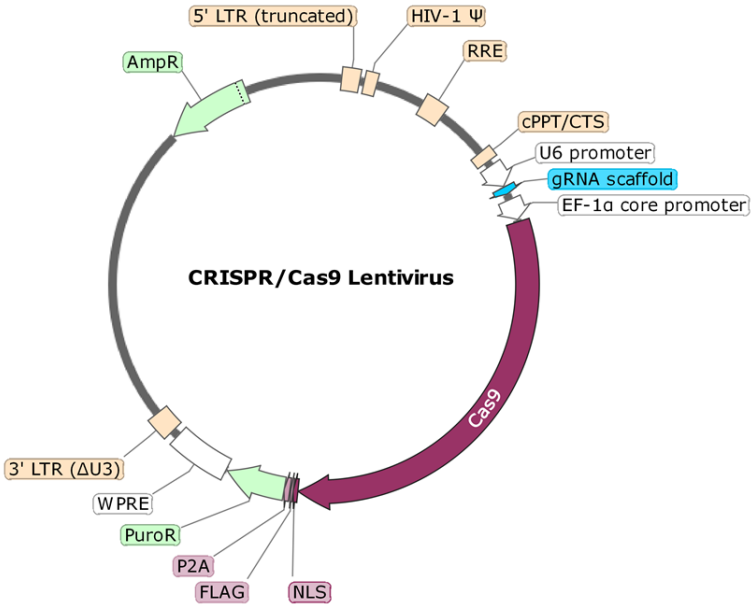

Figure 1: Schematic of the lenti-vector used to generate the CIITA CRISPR/Cas9 Lentivirus.

Gene Target:	sgRNA Sequence:
CIITA	GAGATTCAGGCAGCTCAACG
CIITA	CCATTGCTTGAACCGTCCGG
CIITA	ACATCAAAGTACCCTACAGG
CIITA	GGACAGCTCAAATAGGGCGT
CIITA	GATATTGGCATAAGCCTCCC

Table 1: List of sgRNA Sequences in the CIITA CRISPR/Cas9 Lentivirus.

Application(s)

- Transient knockdown of CIITA in target cells

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 µl x 2) of lentivirus at a titer ≥1 x 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 the SIN (self-inactivation) lentivector which ensures self-inactivation of the lentiviral construct after transduction and after 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.

Figures and Validation Data

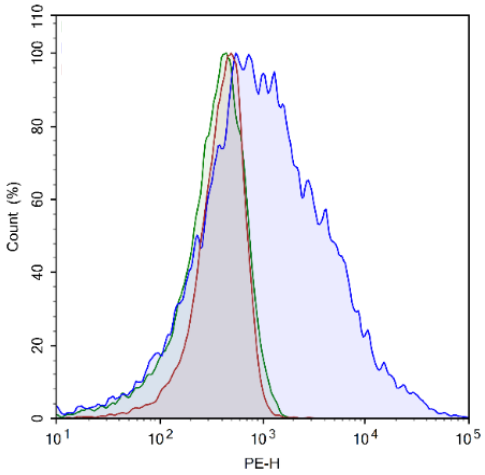

Figure 3: Knockdown of CIITA in THP-1 cells using CIITA CRISPR/Cas9 Lentivirus. 1 x 10^5 THP-1 cells were transduced via spinoculation with 1 x 10^7 TU of CIITA CRISPR/Cas9 lentivirus, corresponding to an MOI >100. 48 hours after transduction, cells were stained with PE-labeled anti-human HLA-DR antibody (R&D Systems, #FAB4869P-100) and analyzed by flow cytometry. Unstained parental THP-1 cells (green) and non-transduced parental THP-1 cells (blue) were compared with transduced THP-1 cells (red). The Y-axis is the % cell number. The X-axis is the intensity of PE.

Troubleshooting Guide

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

License Disclosure

The CRISPR/CAS9 technology is covered under numerous patents, including U.S. Patent Nos. 8,697,359 and 8,771,945, as well as corresponding foreign patents applications, and patent rights.

Related Products

Products	*Catalog #*	*Size*
CIITA CRISPR/Cas9 Lentivirus (Integrating)	78435	500 µl x 2
CIITA Knockout THP-1 Cell Line	78390	2 Vials
B2M/CIITA Double Knockout THP-1 Cell Line	78391	2 Vials
CTLA4 CRISPR/Cas9 Lentivirus (Non-Integrating)	78061	500 µl x 2
CTLA4 CRISPR/Cas9 Lentivirus (Integrating)	78054	500 µl x 2
TIGIT CRISPR/Cas9 Lentivirus (Non-Integrating)	78065	500 µl x 2
TIGIT CRISPR/Cas9 Lentivirus (Integrating)	78058	500 µl x 2
CD47 CRISPR/Cas9 Lentivirus (Non-Integrating)	78063	500 µl x 2
CD47 CRISPR/Cas9 Lentivirus (Integrating)	78056	500 µl x 2
PD-L1 CRISPR/Cas9 Lentivirus (Integrating)	78057	500 µl x 2
PD-L1 CRISPR/Cas9 Lentivirus (Non-Integrating)	78064	500 µl x 2
LAG3 CRISPR/Cas9 Lentivirus (Non-Integrating)	78060	500 µl x 2
LAG3 CRISPR/Cas9 Lentivirus (Integrating)	78053	500 µl x 2