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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

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Data Sheet

CD47 CRISPR/Cas9 Lentivirus (Integrating)

Catalog #: 78056

Description

CD47 (also known as Rh-associated protein, GP42, Integrin-Associated Protein (IAP), or Neurophilin) is an immunoglobulin-like protein that interacts with its receptor, Signal-regulatory protein alpha (SIRPα), on macrophages. This binding interaction regulates transmigration, oxidative burst cytokine production, and phagocytosis, generating a "don't eat me" signal. CD47 is ubiquitously expressed on the surface of normal cells, but is overexpressed in numerous cancer cells where it is thought to contribute to the resistance of tumors to phagocyte-dependent clearance.

The CD47 CRISPR Lentiviruses are replication incompetent, HIV-based VSV-G pseudo-typed lentiviral particles that are ready to be transduced into 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 4 sgRNA (single guide RNA) targeting human CD47 (NM_198793.2) driven by a U6 promoter (Figures 1 and 2).

The integrating lentivirus integrates randomly into the cell's genome to express both the Cas9 and sgRNA. Puromycin selection increases the knockout efficiency by forcing high expression levels of both Cas9 and the sgRNA, and can be used with the integrating lentivirus to quickly and easily achieve high knockdown efficiencies in a cell pool. Efficiencies also depend on the cell type and the gene of interest.

Application

1. Transient knock-down of CD47 in a target cell pool.
2. Generation of a stable CD47 knock-out cell line following limited dilution.

Formulation

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

Titer

Two vials (500 µl x 2) of lentivirus at a titer ≥1 x 10^6 TU/ml. The titer will vary with each lot; the exact value is provided with each shipment.

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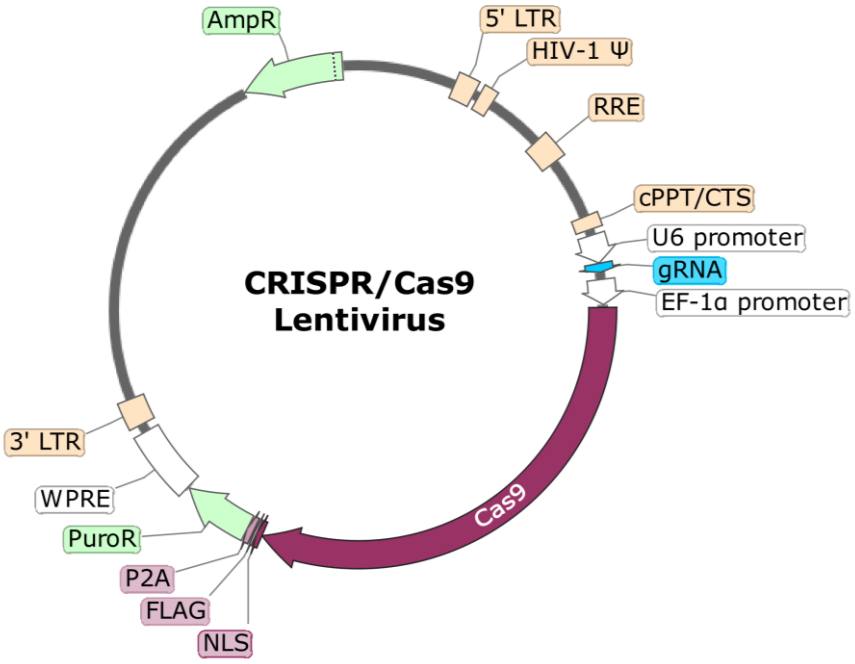

Figure 1. Schematic of the Lenti-vector used to generate the CD47 CRISPR/Cas9 Lentivirus.

Gene Target:	Primer ID:	sgRNA Sequence:
CD47	CD47-1	ATCGAGCTAAAATATCGTGT
CD47	CD47-2	GCACTTAAATATAGATCCGG
CD47	CD47-3	AGTGATGCTGTCTCACACAC
CD47	CD47-4	TTTGCACTACTAAAGTCAGT

Figure 2. List of sgRNA Sequences in the CD47 CRISPR/Cas9 Lentivirus.

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

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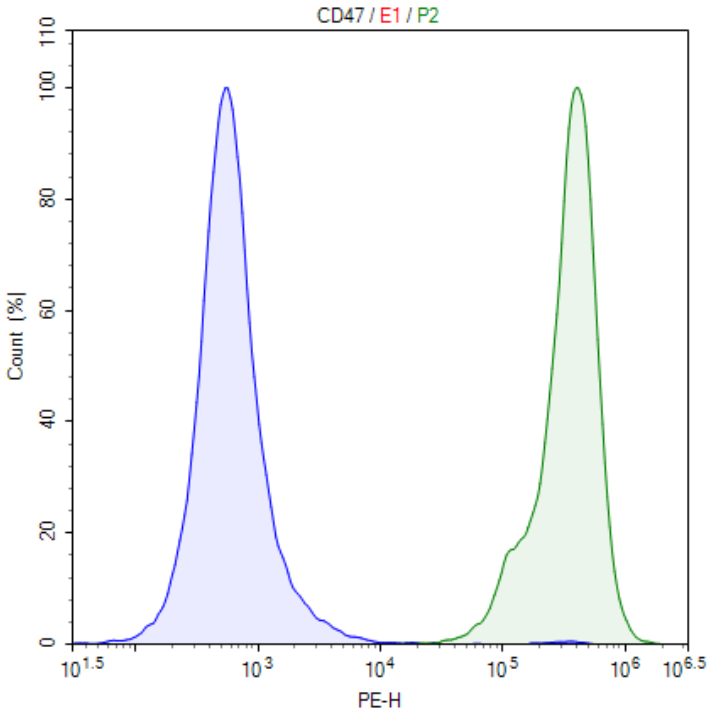

Figure 3. Knock-down of CD47 in CD47 Over-Expressing HEK293 cells.
CD47 Over-expressing HEK293 cells (BPS Bioscience, #71249) were transduced via spinoculation with 5,000,000 TU/well of CD47 CRISPR/Cas9 lentivirus. 72 hours after transduction, cells were stained with PE anti-human CD47 antibody (BioLegend, #323108) and analyzed by FACS. Parental CD47 Over-expressing HEK293 cells are shown in green, and the transduced cells are shown in blue.

Related Products

Product	Cat. #	Size
CD47 CRISPR/Cas9 Lentivirus (Non-Integrating)	78063	500 µl x 2
TCR CRISPR/Cas9 Lentivirus (Integrating)	78055	500 µl x 2
TCR CRISPR/Cas9 Lentivirus (Non-Integrating)	78062	500 µl x 2
Cas9, His-tag (S. pyogenes)	100206-1	50 µg
TCR Knockout NFAT-Luciferase Reporter Jurkat Cell Line	79887	2 vials
CD47 - HEK293 Cell Line	71249	2 vials
Anti-CD47 Antagonist Antibody	79065-1	50 µg
SIRP-α / HEK293 Recombinant Cell Line	60689	2 vials

Notes
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.