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Produktinformation

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- Laborgeräte & Service

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Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Description

NK Viral Transduction Enhancer is a preformulated non-toxic transduction enhancer composed of components A and B, designed to enhance primary NK cell transduction efficiency with lentiviruses.

Background:

Transduction is defined as the process by which foreign DNA is introduced into a cell by a virus or a viral vector. This method of DNA delivery was discovered in 1952 in *Salmonella* and transduction with viral vectors is now an essential tool in molecular biology. Four different types of viral vectors are commonly used: retrovirus, lentivirus, adenovirus and adeno-associated virus. The use of transduction enhancers is common practice as a method to increase efficiency, particularly in cell types known to be hard to transduce, such as primary cells, by improving the cell-virus contact. Transduction of primary NK cells is notoriously challenging, requiring extensive protocol optimization. Their resistance to transduction has been a major handicap in the development of NK-based immunotherapies, particularly at large scale. Optimization efforts have been focusing on the use of specific lentivectors, such as baboon envelope pseudotyped lentivectors, and feeder cells. An easy to use, scalable solution brings major benefits to the manufacture of CAR (chimeric antigen receptor)-NK cells.

Applications:

Lentiviral-mediated primary transduction.

Supplied Materials

Name	Amount	Storage
1000 X NK Viral Transduction Enhancer Component A	100 µl	-20°C
1000 X NK Viral Transduction Enhancer Component B	100 µl	-20°C

Materials Required but Not Supplied

These materials are not supplied with NK Viral Transduction Enhancer but are necessary to follow the protocol described in the "Validation Data" section. Media and reagents used at BPS Bioscience are all validated and optimized for use with this product and are highly recommended for best results.

Name	Ordering Information
Expanded Human Peripheral Blood NK Cells, Frozen	BPS Bioscience #78798
NK Medium, Serum-Free	BPS Bioscience #82615
NK Cell Culture Cytokine Cocktail	BPS Bioscience #82616
Growth-Arrested NK Feeder Cells	BPS Bioscience #78912
Anti-CD19 CAR Lentivirus (CD19 ScFv-CD8-4-1BB-CD3ζ; SIN Vector)	BPS Bioscience #78601
6-well tissue culture plate	

Storage

Store at -20°C for up to 2 years.

Protocol

- The following protocol is a general guideline for transducing primary NK cells using Anti-CD19 CAR Lentivirus (CD19 ScFv-CD8-4-1BB-CD3ζ; SIN Vector) (#78601). The optimal transduction conditions (e.g. MOI, time of assay development) may need to be optimized according to the assay requirements.
- Optimal MOI and transduction efficiency of primary NK cells can be donor dependent.

Day 1:

1. Thaw frozen NK cells according to the protocol in the "Cell Thawing and Culture Protocol" section of the datasheet for #78798 using NK Medium, Serum-Free (#82615) supplemented with 1x NK Cell Culture Cytokine Cocktail (#82616). Add Growth-Arrested NK Feeder Cells (#78912) to the NK cells at a ratio of 1:1 and grow the cells in a 5% CO_2 incubator at 37°C for 3 days before lentiviral transduction.

Day 3:

1. Harvest NK Cells by centrifugation at 300 *x g* for 5 minutes and resuspended in completed NK Medium, Serum-Free at 0.1-0.2 x 10^6 cells/ml.

2. Add 1000x NK Viral Transduction Enhancer Component A and B to the cells to have a 1x final concentration of component A and B in the cell suspension and incubate for 30 minutes at Room Temperature (RT).

3. Transduce the cells with Anti-CD19 CAR Lentivirus (#78601) with the pre-determined optimal MOI in the presence of 1x of NK Viral Transduction Enhancer, by spinoculation at 400 *x g* for 2 hours.

4. Transfer the transduced cells to a tissue culture plate, incubate at 37°C with 5% CO_2 for 6 hours, remove the virus by refreshing the completed NK Medium.

Day 4:

1. Repeat the lentiviral transduction steps (step 1-4 from Day 3).

2. Culture and expand CAR-NK cells and non-transduced NK cells in complete NK Medium.

Day 7:

1. NK cells are ready for downstream analysis.

Validation Data

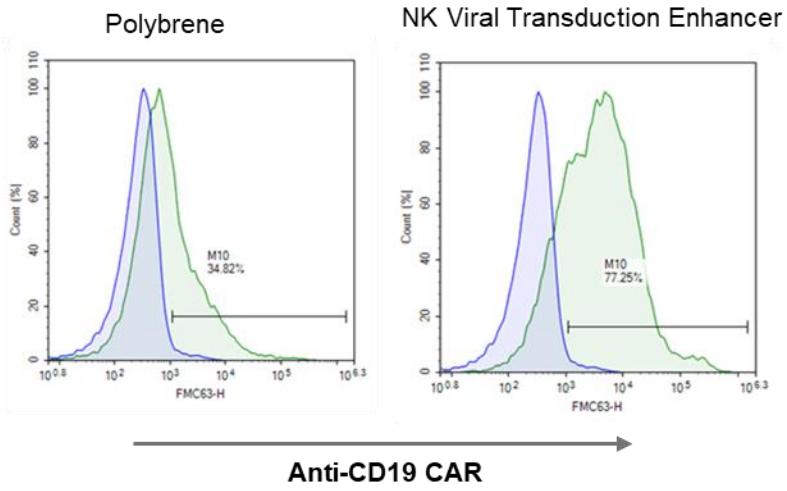

Figure1. NK Viral Transduction Enhancer results in better transduction efficiency than polybrene in Anti-CD19 CAR- NK generation.
Expanded Human Peripheral Blood NK Cells (#78798) were transduced with Anti-CD19 CAR Lentivirus (#78601) at a MOI of 40, in presence of either 8 µg/ml Polybrene (left) or 1x NK Viral Transduction Enhancer (right). 72 hours post-transduction anti-CD19 CAR expression was analyzed by flow cytometry using PE-anti-FMC63 ScFv (Acrobiosystems #FM3-HPY53-25tests). Non-transduced NK cells were used as negative control. The y axis corresponds to the cell %, while the x axis represents the fluorophore intensity.

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

References
Colamartino A., *et al.*, 2019 *Front Immunol.* 10:2873.
Allan D., *et al.*, 2021 *Mol Ther Methods Clin Dev.* 20:559-571.

Related Products

Products	*Catalog #*	*Size*
NK Cell Expansion Kit	78927	1 Kit
Human NK Cell Isolation Kit	82287	1 x 10^8
NCAM/CD56 Positive Cell Isolation Kit	78808	1 x 10^8/1 x 10^9
Anti-NCAM1 Antibody, FITC-Labeled	101865	25 µg/100 µg
IFN-γ (Human) Colorimetric ELISA Detection Kit	79777-1	96 reactions/5 x 96 reactions
Cytotoxicity Dye Kit (CSFE, 7-AAD)	82296	1 kit

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