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Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Description

B2M HLA-A*01:01 Lentivirus are replication incompetent, HIV-based, VSV-G pseudotyped lentiviral particles ready to transduce most mammalian cells, including primary and non-dividing cells. These viruses result in the expression of B2M (beta-2 microglobulin) HLA (human leukocyte antigen)-A*01:01 driven by an EF1a promoter. The lentiviruses also contain a puromycin selection marker (Figure 1).

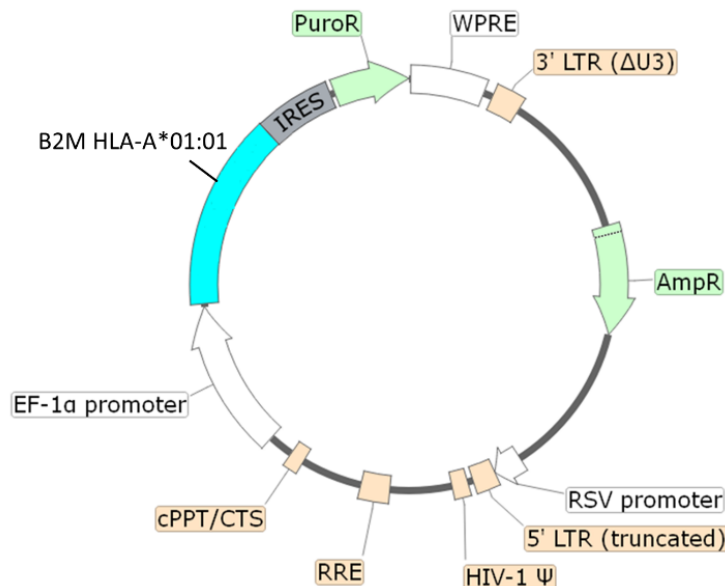


Figure 1. Schematic of the lenti-vector used to generate B2M HLA-A*01:01 Lentivirus.

Background

Human Leukocyte Antigen-A (HLA-A) is an MHC-I (major histocompatibility complex) heavy chain receptor, composed of HLA-A and β 2-microglobulin (B2M). There are over 200 genes encoding HLA variants and this variability plays a critical role in adaptive immunity. HLA-A*01:01 is more common in Europe. It has been broadly linked to autoimmune disorders, type 1 diabetes, and infectious diseases. The presence of this haplotype was considered a risk for the development of severe COVID19, while it was also reported that it had a protective effect against severe bilateral pneumonia. Studies in third wave patients pointed to an effect in forming long-term T cell immunity. Further in-depth studies are needed to fully understand the role of this haplotype as allele of risk or protection in immune-related disorders.

Application(s)

- Expression of human B2M HLA-A*01:01 in cells of interest.
- Generate B2M HLA-A*01:01 expressing cell pools or stable cell lines by puromycin selection.

Formulation

The lentivirus particles were produced in HEK293T cells. They are supplied in medium containing 90% DMEM + 10% FBS. Virus particles can be packaged in custom formulations and produced at higher titers by special request, for an additional fee.

Size and 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 a 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.

Notes

To generate an HLA-A*01:01 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, <https://bpsbioscience.com/kill-curve-protocol>), for antibiotic selection of transduced cells, followed by clonal selection.

Validation Data

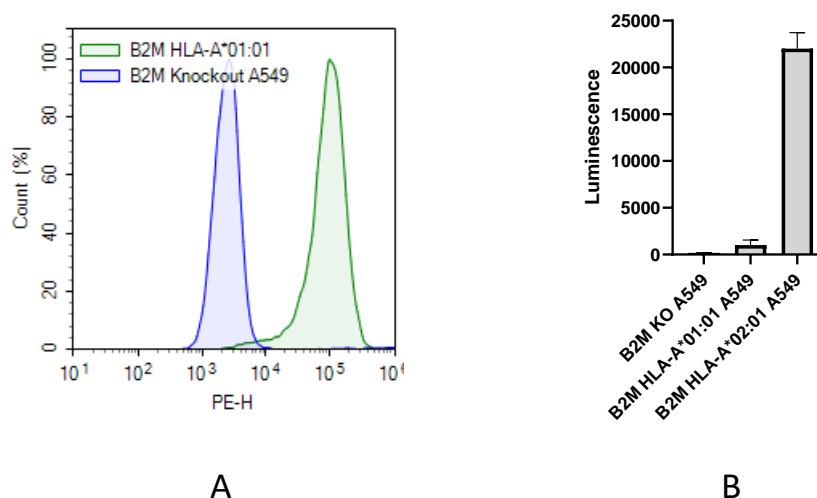


Figure 2. Expression and function of HLA-A*01:01 in B2M Knockout A549 cells transduced with HLA-A*01:01 Lentivirus.

A. Approximately 100,000 B2M Knockout A549 cells (#82871) were transduced with 1×10^6 TU ($100 \mu\text{l}$ of 10^7 TU/ml) of HLA-A*01:01 Lentivirus via spinoculation ($800 \times g$ at 32°C for 30 minutes) in the presence of $5 \mu\text{g/ml}$ of Lenti-Fuse™ Polybrene Viral Transduction Enhancer (#78939). 48 hours post-transduction, the cells were stained with PE anti-human HLA-ABC Antibody (Biolegend #311406) and analyzed by flow cytometry. The y-axis represents the cell % and the x-axis indicates PE intensity.

B. B2M Knockout A549 cells (#82871) were transduced with B2M HLA-A*01:01 Lentivirus or B2M HLA-A*02:01 Lentivirus (#82424) and were loaded with 10 nM NY-ESO-1 peptide (p157-165; #78758), and cocultured with NY-ESO-1 TCR (1G4) CD8^+ NFAT Luciferase Reporter Jurkat Cell Line (#78769) overnight. Luciferase activity was measured with ONE-Step™ Luciferase Assay System (#60690). The results are shown as raw luminescence readings. The NY-ESO-1 TCR (1G4) is a specific T cell receptor that recognizes the NY-ESO-1 epitope presented by the HLA-A*02:01 molecule. B2M knockout A549 cells were run in parallel as a negative control.

Data is representative.

References

Gornalusse G., *et al.*, 2017 *Nature Biotechnology* 35:765-772.
Shkurnikov M., *et al.*, 2023 *PeerJ* 11:e14707.

Troubleshooting Guide

Visit bpsbioscience.com/lentivirus-faq for detailed troubleshooting instructions. For lot-specific information and all other questions, please visit <https://bpsbioscience.com/contact>.

Related Products

<i>Products</i>	<i>Catalog #</i>	<i>Size</i>
HLA-A*02:01 Lentivirus	82424	500 µl x 2
B2M Knockout A549 Cell Line	82871	2 vials
HLA-C*08:02 K562 Cell Line	78974	2 vials
HLA-C*08:02 Lentivirus	78930	500 µl x 2
HLA-A/B/C Knockout Electroporation Kit	82395	1 Kit
HLA-A/B/C Knockout HEK293T Cell Line	82943	2 vials

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