

Description

Beta-2 Microglobulin (B2M) is a required component of Major Histocompatibility Complex (MHC) class 1 molecules, which present peptide fragments from within the cell to cytotoxic T-cells as part of the adaptive immune system.

The B2M 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 B2M driven by a U6 promoter (Figures 1 and 2).

The integrating lentivirus integrates randomly into the cellular genome to express both Cas9 and the sgRNA. Puromycin selection forces 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 will depend on the cell type and the gene of interest.

Application

1. Transient knock-down of B2M in target cells
2. Generation of a stable B2M knock-out cell pool following puromycin selection and limited dilution

Formulation

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

Titer

Two vials (500 μ l x 2) of lentivirus at a titer $\geq 1 \times 10^6$ 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



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

License Disclosure

Visit bpsbioscience.com/license for the label license and other key information about this product.

Troubleshooting Guide

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

Figures and Validation Data

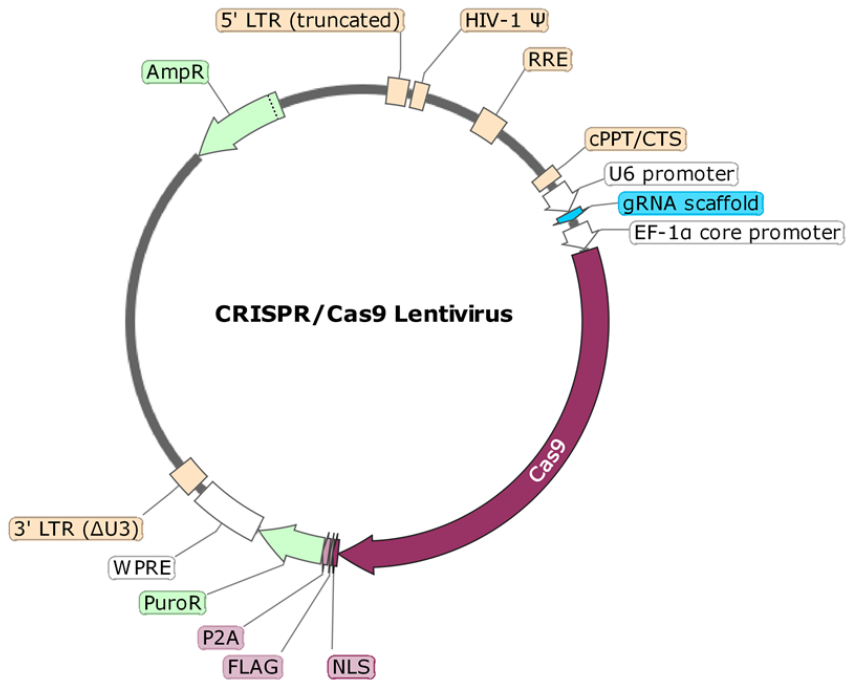


Figure 1. Schematic of the Lenti-vector used to generate the B2M CRISPR/Cas9 Lentivirus

Gene Target:	sgRNA Sequence
B2M	AAGTCAACTTCAATGTCGGA
B2M	CTGAATCTTTGGAGTACCTG
B2M	GAGTAGCGCGAGCACAGCTA
B2M	TCCTGAATTGCTATGTGTCT
B2M	GAAGTTGACTTACTGAAGAA

Figure 2. List of sgRNA Sequences in the B2M CRISPR/Cas9 Lentivirus

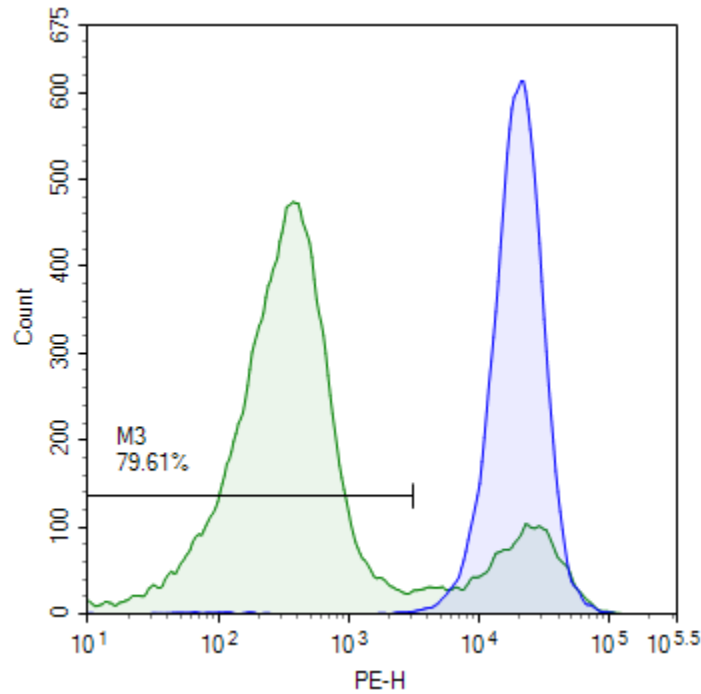


Figure 3. Knock-down of B2M in Jurkat cells.

Parental Jurkat cells were transduced via spinoculation with B2M CRISPR/Cas9 lentivirus. 24 hours after transduction, cells were selected for another 24 hours with puromycin, then stained with PE-labeled anti-human B2M antibody (BioLegend, #395703) and analyzed by flow cytometry. Parental Jurkat cells are shown in blue, and the transduced cells are shown in green.

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.

Related Products

<i>Products</i>	<i>Catalog #</i>	<i>Size</i>
B2M (Human) CRISPR/Cas9 Lentivirus (Non-Integrating)	78341	500 µl x 2
FcRn (FCGRT/B2M), His-Tag (Mouse) HiP™	11349	100 µg
FcRn (FCGRT/B2M), His-Tag	71285	100 µg/1 mg
FcRn (FCGRT/B2M), His-Tag, Biotin-Labeled, (Human) HiP™	71283	25 µg/50 µg
FcRn (FCGRT/B2M), His-Avi-Tag, Biotin-Labeled (Mouse)	71286	50 µg