

**Description**

Transforming growth factor beta receptor 2 (TGFBR2) encodes the TGF-β receptor protein, a transmembrane protein that forms a heterodimeric complex with other receptor proteins and binds TGF-β. This receptor/ligand complex phosphorylates proteins which regulate cell proliferation, cell cycle arrest, wound healing, and immunosuppression. Mutations in TGFBR2 have been linked with Marfan syndrome and the development of various types of tumors.

The TGFBR2 CRISPR/Cas9 Lentiviruses are replication incompetent, HIV-based VSV-G pseudotyped lentiviral particles ready to infect most 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 RNA) targeting human TGFBR2 (Figure 1 and Table 1).

The TGFBR2 CRISPR/Cas9 non-integrating lentivirus is made with a mutated integrase, resulting in only transient expression of Cas9 and sgRNA. Although using the non-integrating lentivirus results in lower knockdown efficiency, the Cas9 protein is not permanently expressed, which lowers the risk of off-targeting, and there are no random integrations into the cell’s genome. Despite transient expression of Cas9 and sgRNA, knockout cell lines can still be generated using cell sorting or limiting dilution due to the permanent changes in the genomic DNA from the Cas9 nuclease activity and NHEJ repair.

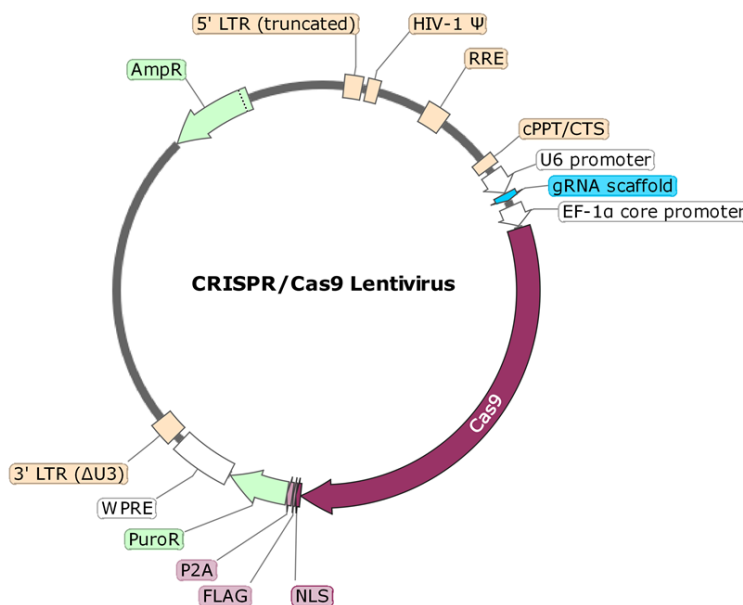


Figure 1: Schematic of the lenti-vector used to generate the TGFBR2 CRISPR/Cas9 Lentivirus.

Gene Target:	sgRNA Sequence:
TGFBR2-1-F	ACAGTGATCACACTCCATGT
TGFBR2-2-F	TATCATGTCGTTATTAAC TG
TGFBR2-3-F	GCAGAAGCTGAGTTCAACCT
TGFBR2-4-F	AAAGCGACCTTTCCCCACCA
TGFBR2-5-F	ACCTACAGGAGTACCTGACG

Table 1: List of sgRNA Sequences in the TGFBR2 CRISPR/Cas9 Lentivirus.

### Application(s)

- Transient knockdown of TGFBR2 in target cell pools
- Generation of a stable TGFBR2 knockout cell line following puromycin selection and limiting dilution cloning

### 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  $\mu$ l x 2) of lentivirus at a titer  $\geq 1 \times 10^7$  TU/ml. The titer will vary with each lot; the exact value is provided with each shipment.

### Storage



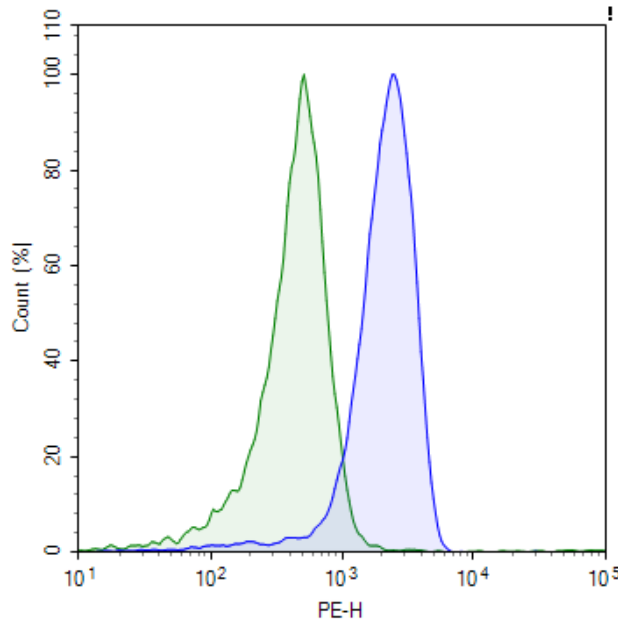
Lentiviruses are shipped with dry ice. For long-term storage, it is recommended to store the lentiviruses at  $-80^{\circ}\text{C}$ . Avoid repeated freeze-thaw cycles. Titers can drop significantly with each freeze-thaw cycle.

### 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.

**Validation Data**



*Figure 2: Knockdown of TGFBR2 in Jurkat cells using TGFBR2 CRISPR/Cas9 Lentivirus.* Jurkat cells were transduced via spinoculation with  $1 \times 10^7$  TU/well of TGFBR2 CRISPR/Cas9 lentivirus, corresponding to an MOI >5. 48 hours after transduction, cells were stained with TGFBR2 Recombinant Polyclonal antibody (ThermoFisher #710276) followed by a PE-labeled anti-rabbit secondary (BioLegend #406421) and analyzed by flow cytometry. Non-transduced, parental Jurkat cells are shown in blue, and the transduced cells are shown in green.

**Troubleshooting Guide**

Visit [bpsbioscience.com/lentivirus-faq](https://bpsbioscience.com/lentivirus-faq) for detailed troubleshooting instructions. For all further questions, please email [support@bpsbioscience.com](mailto:support@bpsbioscience.com).

**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>
CTLA4 CRISPR/Cas9 Lentivirus (Non-Integrating)	78061	500 $\mu$ l x 2
CTLA4 CRISPR/Cas9 Lentivirus (Integrating)	78054	500 $\mu$ l x 2
TIGIT CRISPR/Cas9 Lentivirus (Non-Integrating)	78065	500 $\mu$ l x 2
TIGIT CRISPR/Cas9 Lentivirus (Integrating)	78058	500 $\mu$ l x 2
CD47 CRISPR/Cas9 Lentivirus (Non-Integrating)	78063	500 $\mu$ l x 2
CD47 CRISPR/Cas9 Lentivirus (Integrating)	78056	500 $\mu$ l x 2