

Data Sheet

Simplicon® B18R RNA (human codon optimized)

Synthetic RNA

SCR723**Pack Size: 10 µg****Store at -80 °C****FOR RESEARCH USE ONLY****Not for use in diagnostic procedures. Not for human or animal consumption.**

Background

Simplicon® RNA is a novel system to effect immediate high sustained protein expression of multiple genes into transfected cells without the risk of genome integration. The technology employs a single, synthetic, polycistronic, self-replicating RNA based on the Venezuelan equine encephalitis (VEE) genome.^{1,2,3} Simplicon® RNA contains only genes encoding the VEE RNA replication machinery while the structural proteins that are required to make an infectious particle have been removed and replaced with the transgenes of interest.

Introduction and replication of Simplicon® RNA is expected to elicit a strong interferon response in transfected cells. To suppress the IFN responses, a Vaccinia virus protein4, B18R, is used for the original Simplicon® RNA technology.

The B18R RNA (human codon optimized) is a synthetic mRNA that is co-transfected with the Simplicon® RNA to suppress the IFN responses. B18R RNA can also be used to prepare B18R conditioned medium by transfection into HFFs (SCC058). B18R RNA can also be used for co-transfection of any mRNAs to suppress the IFN responses and enable repeated transfection of mRNAs. Please refer to the User Guide for the Simplicon® Expression System located at SigmaAldrich.com for detailed protocols.

Quality Control Testing

- Discrete RNA band on RNA gel of appropriate size.
- 30-60% GFP-positive human foreskin fibroblasts (HFFs) with FACS analysis

Storage and Handling

- B18R RNA (human codon optimized): (CS224516) One (1) vial containing 10 µL of RNA (1 µg/µL).
- Store at -80 °C.
- For best recovery, quick spin the vials after thawing on ice. Do not vortex. Aliquot into sterile, nuclease-free Eppendorf® tubes on ice and store at -80 °C. Limit repeated freeze-thaw cycles.

Representative Data



Figure 1. 5' capped RNA was *in vitro* transcribed. The poly (A) tail was added by an enzymatic poly (A) adenylation reaction. cDNA for B18R was human codon optimized.

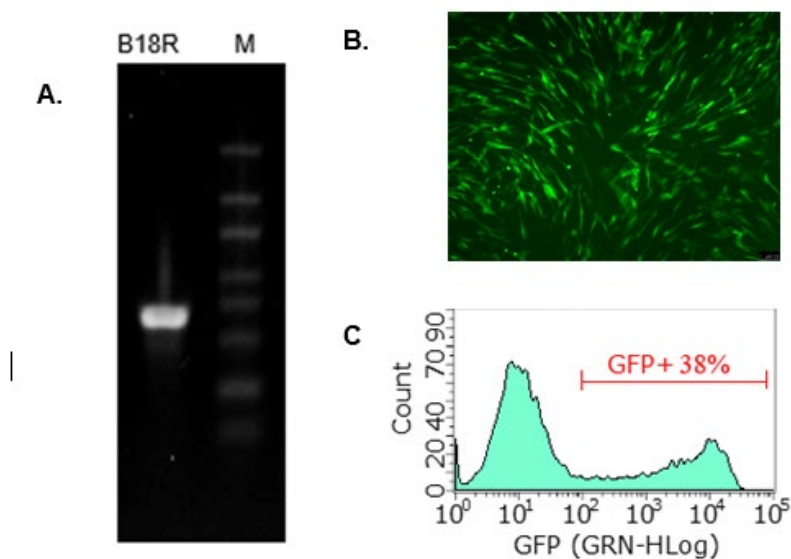


Figure 2. RNA gels and TagGFP2 expression in HFFs. **(A)** B18R RNA gel. Marker: 6, 4, 3, 2, 1.5, 1, 0.5, 0.2 kb **(B, C)** TagGFP2 Simplicon® RNA (E3L) and B18R RNA were co-transfected into HFFs with Messenger Max™ transfection Reagent. One day after transfection, GFP expression was analyzed by fluorescence microscopy and FACS.

Related Products

Product catalog numbers indicated in () can be purchased at SigmaAldrich.com unless otherwise stated.

Cat No.	Description
SCR720	TagGFP2 Simplicon® RNA (E3L) Kit
SCR721	TagRFP Simplicon® RNA (E3L) Kit
SCR722	B18R-E3L RNA (human codon optimized for B18R and E3L)
SCR724	Simplicon® Cloning Vector (E3L)
SCR725	TagGFP2 Simplicon® Plasmid (E3L)
SCR726	TagRFP Simplicon® Plasmid (E3L)
SCR727	B18R-E3L Plasmid (human codon optimized for B18R and E3L)
SCR728	B18R Plasmid (human codon optimized)
GF156	B18R protein (produced from insect cells)
GF197	B18R protein (produced from HEK 293 cells)

References

1. Yoshioka, N., et. al. Cell Stem Cell 2013, 13(2):246-254.
2. Yoshioka N., and Dowdy SF. PLoS One. 2017;12(7):e0182018. doi: 10.1371/journal.pone.0182018. eCollection 2017.
3. Pegg G., et al. Drug Metabolism and Pharmacokinetics, Volume 33, Issue 1, Supplement, 2018; Pages S33-S34
4. Perdiguero B., and Esteban M. J Interferon Cytokine Res. 2009;29(9):581-98. doi: 10.1089/jir.2009.0073.

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Document Template 20306518 Ver 6.0

20327666 Ver 4.0, Rev 14JUN2024, LB

