

Control Lentiviral shRNA Construct

Shipment Contents: Control Lentiviral shRNA Construct (Plasmid) shNT in pRSIT17-U6Tet-sh-CMV-TetRep-2A-TagGFP2-2A-Puro — Store at -20°C

Description:

Cellecta's shRNA constructs are lentiviral vectors expressing short-hairpin RNA for knockdown of target mRNA transcripts. The use of our proprietary shRNA design algorithm and database of validated shRNAs results in a high percentage of functional sequences, typically at least 70% knockdown efficiency for approximately 70% of the shRNA constructs. Results may vary between different cell types.

The non-targeting (NT) lentiviral shRNA construct expresses an shRNA sequence with no significant homology to any mammalian transcript.

The plasmid shRNA construct can be packaged into VSV-G pseudotyped viral particles using most commerciallyavailable second or third-generation packaging mixes or Cellecta's second-generation psPAX2/pMD2.G packaging plasmid mix:

• Cat.# CPCP-K2A, Ready-to-use Lentiviral Packaging Plasmid Mix, 250 µg (for 25 x 10-cm plates)

The titer of packaged constructs can be functionally determined by transduction of 293T cells and either FACS of RFP- or GFP-positive cells, antibiotic selection assay, or by PCR titering of integrated viral DNA.

Biosafety Level:	BSL-2
Storage:	-20°C
Shelf Life:	2 years from date of receipt
Shipping Conditions:	Room Temperature, Blue Ice, or Dry Ice

Product Information (Cellecta Website):

 User Manual:
 https://www.cellecta.com/product-manuals-and-certificates/

 Vector Info (Sequence, etc.):
 https://www.cellecta.com/vector-information/

Control Lentiviral shRNA Construct shNT in pRSIT17-U6Tet-sh-CMV-TetRep-2A-TagGFP2-2A-Puro Cat.# SHCTL-NT-pRSIT17



Contents:

Catalog #	Description
SHCTL-NT-pRSIT17	Control Lentiviral shRNA Construct shNT in pRSIT17-U6Tet-sh-CMV-TetRep-2A-TagGFP2-2A-Puro (plasmid) RefSeg#: n/a, Gene ID: n/a
	<u>Plasmid, >25 μg:</u> 25 μg , 0.5 μg/μl (50 μl × 1 vial) Lot# 17102508; Store at -20°C
Target Sequence:	5'-CAACAAGATGAAGAGCACCAA-3'
shRNA Insert:	ACCGGCAACAAGATGGAGAGCACTAAGTTAATATTCATAGCTTGGTGCTCTTCATCTTGTTGTTTT
Sequencing QC:	ANNTTCTTGGGNAGTTTGCAGTTTTAAAATTATGTTTTAAAATGGACTATCATATGCTTACCGTAACTTGAAAGCCCTAT CAGTGATAGAGATTTATATATCCCTATCAGTGATAGAGACACCGGCAACAAGATGGAGAGCACTAAGTTAATATTCATAG CTTGGTGCTCTTCATCTTGTTGTTTTTTCGGACTGTAGAACTCTGAACCTCGAGCAATTTAAAAGAAAAGGGGGGGATTGG GGGGTACAGTGCAGGGGAAAGAATAGTAGACATAATAGCACAGACATACAA
Sequencing Primer:	5'-ATTAGTACAAAATACGTGACGTAGAA-3' (U6-3)

Design of Cellecta shRNA Constructs:

U6><u>ACCG</u>G-21mer(**sense** w/ mismatch(es)*)-GTTAATATTCATAGC(**loop**)-21mer(**antisense**)-TTTT<u>TTCG</u>

ACCG/TTCG : Cloning Sites

* For some constructs, the sense portion of the shRNA may contain specific mismatches introduced by Cellecta. We incorporate some mismatches in our library hairpins to improve stability and representation in a pooled-screening format. Although we have not observed any significant differences in efficacy between perfectly homologous shRNA sequences and those with 1-2 non-sequential mismatches, we typically use the same hairpin design for individual constructs to maintain sequence consistency with our functionally active library designs. Note that this design may not apply in the case of customer-requested designs.

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