

Control shRNA Lentiviral Particles-A: sc-108080

BACKGROUND

RNA interference (RNAi) is one of the most exciting discoveries of the past decade in functional genomics and proteomics. While first recognized in nematodes as a response to exogenously introduced long double-stranded RNA (dsRNA), it is now clear that RNAi is utilized by most eukaryotes *in vivo* for anti-viral defense, transposon activity modulation and gene regulation, and has rapidly become an important research tool for gene silencing. Specifically, RNAi is the pathway by which short interfering RNA (siRNA) or short hairpin RNA (shRNA) are used to silence the expression of target genes. Compared to siRNA, shRNA offers advantages in silencing longevity and delivery.

Upon introduction, the shRNA plasmid DNA enters the cell where shRNA is transcribed. The shRNA is then cleaved by an RNase III-like enzyme called Dicer into small interfering RNA (siRNA), which are short RNA duplexes of 19-21 nucleotides with two nucleotide 3' overhangs on each strand. The siRNAs are then assembled into endoribonuclease-containing complexes known as RNA-induced silencing complexes (RISCs), unwinding in the process. Activated RISCs subsequently bind to complementary transcripts by base pairing interactions between the siRNA anti-sense strand and complementary mRNA. The bound mRNA is cleaved and sequence specific degradation of mRNA results in gene silencing. In mammalian cells, introduction of long dsRNA (more than 30 nucleotides) initiates a potent anti-viral response, exemplified by nonspecific inhibition of protein synthesis and RNA degradation. The mammalian anti-viral response can be bypassed, however, by the introduction of siRNAs or shRNA plasmid DNA.

Santa Cruz Biotechnology, Inc. currently offers more than 49,000 target specific shRNA Lentiviral Particles that encode 19-25 nucleotide (plus hairpin) shRNAs that can be used to knock down protein expression in a broad variety of mammalian cell types. Our product line includes shRNA lentiviral particles designed to silence a large selection of proteins, including tumor suppressors, transcription regulators, cell cycle proteins, membrane receptors, signaling intermediates, kinases, cell adhesion proteins and proteins involved in lymphocyte signaling. In addition, for each shRNA Lentiviral Particles product, we offer an appropriate "matched" control antibody for confirmation of targeted mRNA silencing by either Western blotting or fluorescence antibody cell staining. We also offer appropriate buffers and non-targeted control shRNA Lentiviral Particles.

PRODUCT

Control shRNA Lentiviral Particles-A is a negative control for experiments using targeted shRNA Lentiviral Particle transduction; Control shRNA Lentiviral Particles encodes a scrambled shRNA sequence that will not lead to the specific degradation of any known cellular mRNA. After transduction, cells stably expressing the control shRNA may be isolated via puromycin selection. Each vial contains 200 μ l shRNA lentiviral particles sufficient for 10-20 transductions.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

shRNA LENTIVIRAL PARTICLES SUPPORT REAGENTS

PRODUCT	CAT. #	DESCRIPTION	AMOUNT
Electrophoresis Sample Buffer	sc-24945	Ready-to-use reducing electrophoresis sample buffer solution for the preparation of protein samples to be separated in SDS-PAGE.	25 ml; 2X concentrate
RIPA Lysis Buffer	sc-24948	For use in mammalian cell lysis; with protease inhibitors. Available in four vials: 1X lysis buffer, PMSF, protease inhibitor cocktail and sodium orthovanadate.	50 ml
Puromycin dihydrochloride	sc-108071	Available for selection and maintenance of cells transfected with the puromycin-N-acetyl-transferase (pac) gene.	25 mg

SELECT PRODUCT CITATIONS

- Pavlov, T.S., et al. 2010. Endothelin-1 inhibits the epithelial Na⁺ channel through β Pix/14-3-3/Nedd4-2. *J. Am. Soc. Nephrol.* 21: 833-843.
- Fritz, R.D., et al. 2010. CNK1 is a novel Akt interaction partner that promotes cell proliferation through the Akt-FoxO signalling axis. *Oncogene* 29: 3575-3582.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.