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Exportin 5 shRNA (m) Lentiviral Particles: sc-45570-V

BACKGROUND

The karyopherin-related nuclear transport factor Exportin 5, also known as Exp5, preferentially recognizes and transports RNAs containing minihelix motifs, structural cis-acting export elements that comprise a double-stranded stem (14 nucleotides) with a base-paired 5' end and a 3-8-nucleotide protruding 3' end. Exportin 5 also mediates protein transport between the nuclear and cytoplasmic compartment. Exportin 5 belongs to a large family of karyopherins and stimulates nuclear export of dsRNA binding proteins eEF1A and tRNA.

REFERENCES

- Bohnsack, M.T., et al. 2002. Exp5 exports eEF1A via tRNA from nuclei and synergizes with other transport pathways to confine translation to the cytoplasm. *EMBO J.* 21: 6205-6215.
- Brownawell, A.M., et al. 2002. Exportin 5, a novel karyopherin, mediates nuclear export of double-stranded RNA binding proteins. *J. Cell Biol.* 156: 53-64.
- Chen, T., et al. 2004. Nucleocytoplasmic shuttling of Jaz, a new cargo protein for Exportin 5. *Mol. Cell. Biol.* 24: 6608-6619.
- Gwizdek, C., et al. 2004. Minihelix-containing RNAs mediate Exportin 5-dependent nuclear export of the double-stranded RNA-binding protein ILF3. *J. Biol. Chem.* 279: 884-891.
- Macchi, P., et al. 2004. The brain-specific double-stranded RNA-binding protein Staufin2: nucleolar accumulation and isoform-specific Exportin 5-dependent export. *J. Biol. Chem.* 279: 31440-31444.

CHROMOSOMAL LOCATION

Genetic locus: Xpo5 (mouse) mapping to 17 C.

PRODUCT

Exportin 5 shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Exportin 5 siRNA (m): sc-45570 and Exportin 5 shRNA Plasmid (m): sc-45570-SH as alternate gene silencing products.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

APPLICATIONS

Exportin 5 shRNA (m) Lentiviral Particles is recommended for the inhibition of Exportin 5 expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Exportin 5 (E-16): sc-34619 is recommended as a control antibody for monitoring of Exportin 5 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Exportin 5 gene expression knockdown using RT-PCR Primer: Exportin 5 (m)-PR: sc-45570-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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.

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.

PROTOCOLS

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