



CRM1 siRNA (m): sc-35117

BACKGROUND

Protein transport across the nucleus is a selective, multistep process involving several cytoplasmic factors. Proteins must be recognized as import substrates, dock at the nuclear pore complex and translocate across the nuclear envelope in an ATP-dependent fashion. Two cytosolic factors centrally involved in the recognition and docking process are the karyopherin α 1 and karyopherin β 1 subunits. p62 glycoprotein is a nucleoporin that is not only involved in the nuclear import of proteins, but also the export of nascent mRNA strands. NTF2 (nuclear transport factor 2) interacts with nucleoporin p62 as a homodimer composed of two monomers, and may be an obligate component of functional p62. CRM1 has been shown to be an export receptor for leucine-rich proteins that contain the nuclear export signal (NES).

REFERENCES

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2. Paschal, B.M., et al. 1995. Identification of NTF2, a cytosolic factor for nuclear import that interacts with nuclear pore complex protein p62. *J. Cell Biol.* 129: 925-937.
3. Dargemont, C., et al. 1995. Direct interaction of nucleoporin p62 with mRNA during its export from the nucleus. *J. Cell Sci.* 108: 257-263.
4. Lounsbury, K.M., et al. 1996. Ran binding domains promote the interaction of Ran with p97/ β -karyopherin, linking the docking and translocation steps of nuclear import. *J. Biol. Chem.* 271: 2357-2360.
5. Moroianu, J., et al. 1996. The binding site of karyopherin α for karyopherin β overlaps with a nuclear localization sequence. *Proc. Natl. Acad. Sci. USA* 93: 6572-6576.
6. Moroianu, J., et al. 1996. Nuclear protein import: Ran-GTP dissociates the karyopherin $\alpha\beta$ heterodimer by displacing α from an overlapping binding site on β . *Proc. Natl. Acad. Sci. USA* 93: 7059-7062.

CHROMOSOMAL LOCATION

Genetic locus: Xpo1 (mouse) mapping to 11 A3.2.

PRODUCT

CRM1 siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CRM1 shRNA Plasmid (m): sc-35117-SH and CRM1 shRNA (m) Lentiviral Particles: sc-35117-V as alternate gene silencing products.

For independent verification of CRM1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-35117A, sc-35117B and sc-35117C.

PROTOCOLS

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

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

CRM1 shRNA Plasmid (m) is recommended for the inhibition of CRM1 expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

CRM1 (C-1): sc-74454 is recommended as a control antibody for monitoring of CRM1 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 reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CRM1 gene expression knockdown using RT-PCR Primer: CRM1 (m)-PR: sc-35117-PR (20 μ l, 422 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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