SANTA CRUZ BIOTECHNOLOGY, INC.

Lamin B1 siRNA (m): sc-35779

BACKGROUND

A unique family of cysteine proteases has been described that differs in sequence, structure and substrate specificity from any previously described protease family. This family, termed CED-3/ICE, function as key components of the apoptotic machinery and act to destroy specific target proteins which are critical to cellular longevity. Nuclear Lamins are critical to maintaining the integrity of the nuclear envelope and cellular morphology as components of the nuclear lamina, a fibrous layer on the nucleoplasmic side of the inner nuclear membrane which is thought to provide a framework for the nuclear envelope and may also interact with chromatin. B-type Lamins, such as Lamin B1, undergo a series of modifications, such as farnesylation and phosphorylation. Lamin B1 is a 586 amino acid protein that is encoded by a gene which, when mutated, is involved in the pathogenesis of autosomal dominant adult-onset leukodystrophy (ADLD), a disease characterized by cerebellar dysfunction and symmetric demyelination of the central nervous system.

REFERENCES

1. Moir, R.D., et al. 1995. The dynamic properties and possible functions of nuclear lamins. Int. Rev. Cytol. 162B: 141-182.
2. Duan, H., et al. 1996. ICE-LAP3, a novel mammalian homologue of the *Caenorhabditis elegans* cell death protein CED-3 is activated during Fas- and tumor necrosis factor-induced apoptosis. J. Biol. Chem. 271: 1621-1625.
3. Duan, H., et al. 1996. ICE-LAP6, a novel member of the ICE/CED-3 gene family, is activated by the cytotoxic T cell protease granzyme B. J. Biol. Chem. 271: 16720-16724.
4. Rao, L., et al. 1996. Lamin proteolysis facilitates nuclear events during apoptosis. J. Cell Biol. 135: 1441-1455.
5. Fernandes-Alnemri, T.F., et al. 1996. *In vitro* activation of CPP32 and Mch3 by Mch4, a novel human apoptotic cysteine protease containing two FADD-like domains. Proc. Natl. Acad. Sci. USA 93: 7464-7469.
6. Takahashi, A., et al. 1996. Cleavage of Lamin A by Mch2 a but not CPP32: multiple interleukin 1 β-converting enzyme-related proteases with distinct substrate recognition properties are active in apoptosis. Proc. Natl. Acad. Sci. USA 93: 8395-8400.

CHROMOSOMAL LOCATION

Genetic locus: Lmnb1 (mouse) mapping to 18 D3.

PRODUCT

Lamin B1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Lamin B1 shRNA Plasmid (m): sc-35779-SH and Lamin B1 shRNA (m) Lentiviral Particles: sc-35779-V as alternate gene silencing products.

For independent verification of Lamin B1 (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-35779A, sc-35779B and sc-35779C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Lamin B1 siRNA (m) is recommended for the inhibition of Lamin B1 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 µM in 66 µl. 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

Lamin B1 (A-11): sc-377000 is recommended as a control antibody for monitoring of Lamin B1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Lamin B1 gene expression knockdown using RT-PCR Primer: Lamin B1 (m)-PR: sc-35779-PR (20 µl, 564 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Fiume, R., et al. 2009. Involvement of nuclear PLC β1 in Lamin B1 phosphorylation and G_2/M cell cycle progression. FASEB J. 23: 957-966.

RESEARCH USE

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