

caspase-6 siRNA (m): sc-72803

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, CED-3/caspase-1, is comprised of caspase-1, caspase-2, caspase-3, caspase-4, caspase-6, caspase-7 (also designated Mch3, ICE-LAP3 or CMH-1), caspase-9 and caspase-10. CED-3/caspase-1 family members function as key components of the apoptotic machinery and act to destroy specific target proteins which are critical to cellular longevity. Poly(ADP-ribose) polymerase plays an integral role in surveying for DNA mutations and double strand breaks. Caspase-3, caspase-7 and caspase-9, but not caspase-1, have been shown to cleave the nuclear protein PARP into an apoptotic fragment. Caspase-6, but not caspase-3, has been shown to cleave the nuclear lamins which are critical to maintaining the integrity of the nuclear envelope and cellular morphology. Caspase-10 has been shown to activate caspase-3 and caspase-7 in response to apoptotic stimuli.

REFERENCES

1. Lindahl, T., et al. 1995. Post-translational modification of poly (ADP-ribose) polymerase induced by DNA strand breaks. *Trends Biochem. Sci.* 20: 405-411.
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. 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.
4. 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.
5. Simbulan-Rosenthal, C.M., et al. 1996. The expression of poly(ADP-ribose) polymerase during differentiation-linked DNA replication complex. *Biochemistry* 35: 11622-11633.

CHROMOSOMAL LOCATION

Genetic locus: Casp6 (mouse) mapping to 3 G3.

PRODUCT

caspase-6 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 caspase-6 shRNA Plasmid (m): sc-72803-SH and caspase-6 shRNA (m) Lentiviral Particles: sc-72803-V as alternate gene silencing products.

For independent verification of caspase-6 (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-72803A, sc-72803B and sc-72803C.

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

caspase-6 siRNA (m) is recommended for the inhibition of caspase-6 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

caspase-6 p10 (H-12): sc-377393 is recommended as a control antibody for monitoring of caspase-6 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:10p00) 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 caspase-6 gene expression knockdown using RT-PCR Primer: caspase-6 (m)-PR: sc-72803-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yao, Y., et al. 2016. Identification of caspase-6 as a new regulator of alternatively activated macrophages. *J. Biol. Chem.* 291: 17450-17466.

RESEARCH USE

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