

caspase-5 siRNA (h): sc-72800

BACKGROUND

Caspases are cysteine proteases which play important roles in the activation of cytokines and in apoptosis. The ICE subfamily of caspases comprises peptides closely related to caspase-1, which promotes maturation of interleukin 1 β (IL-1 β) and interleukin-18 (IL-18) by proteolytic cleavage of precursor forms to generate biologically active peptides. Both caspase-4 and caspase-5 are members of the caspase-1 subfamily, and are more closely related to each other than to other homologues. Caspase-5 (also designated ICErel-III, TY, ICH-3 and caspase-12 in mouse), can cleave its own precursor, an activity that requires the cysteine 245 residue. Frameshift mutations in caspase-5 have been identified in MMP tumors of the endometrium, colon and stomach, indicating the caspase-5 may be a new target gene in the microsatellite mutator pathway for cancer. The human caspase-5 gene maps to chromosome 11q22.3 and encodes a protein whose expression is barely detectable in most tissues except brain, with highest expression levels being found in lung, liver and skeletal muscle.

REFERENCES

1. Munday, N.A., et al. 1995. Molecular cloning and pro-apoptotic activity of ICErelIII and ICErelIII, members of the ICE/CED-3 family of cysteine proteases. *J. Biol. Chem.* 270: 15870-15876.
2. Faucheu, C., et al. 1996. Identification of a cysteine protease closely related to interleukin-1 β -converting enzyme. *Eur. J. Biochem.* 236: 207-213.
3. Cohen, G.M. 1997. Caspases: the executioners of apoptosis. *Biochem. J.* 326: 1-16.

CHROMOSOMAL LOCATION

Genetic locus: CASP5 (human) mapping to 11q22.3.

PRODUCT

caspase-5 siRNA (h) 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-5 shRNA Plasmid (h): sc-72800-SH and caspase-5 shRNA (h) Lentiviral Particles: sc-72800-V as alternate gene silencing products.

For independent verification of caspase-5 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-72800A, sc-72800B and sc-72800C.

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-5 siRNA (h) is recommended for the inhibition of caspase-5 expression in human 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-5 p20 (H-2): sc-393346 is recommended as a control antibody for monitoring of caspase-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 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-5 gene expression knockdown using RT-PCR Primer: caspase-5 (h)-PR: sc-72800-PR (20 μ l, 398 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Zhu, Q., et al. 2012. The role of acid sphingomyelinase and caspase 5 in hypoxia-induced HuR cleavage and subsequent apoptosis in hepatocytes. *Biochim. Biophys. Acta* 1821: 1453-1461.

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.