SANTA CRUZ BIOTECHNOLOGY, INC.

Calpain 9 siRNA (m): sc-62071

BACKGROUND

Calpain 9 belongs to a family of 14 intracellular calcium activated cysteine proteases present in the fungi, plant and animal kingdoms. Several of these proteases have been implicated in cardiovascular diseases. On a high salt diet, Calpain 9 is downregulated by more than 50% in the heart. The differential regulation of Calpain 9 seen under such conditions may play a role in hypertensive target organ damage. The digestive tract-specific Calpain 9 is downregulated in gastric cancer cell lines, suggesting that it acts as a gastric cancer suppressor. Two known isoforms exist for Calpain 9 due to alternative splicing. The two isoforms vary in their amino acid sequences between amino acids 292 and 318.

REFERENCES

1. Murachi, T. 1984. Calcium-dependent proteinases and specific inhibitors: calpain and Calpastatin. Biochem. Soc. Symp. 45: 149-167.
2. Kawasaki, H., et al. 1996. Regulation of the calpain-Calpastatin system by membranes (review). Mol. Membr. Biol. 13: 217-224.
3. Johnson, G.V., et al. 1997. Calpains: intact and active? Bioessays 19: 1011-1018.
4. Huang, Y. and Wang, K.K. 2001. The calpain family and human disease. Trends Mol. Med. 7: 355-362.
5. Markmann, A., et al. 2005. Downregulation of Calpain 9 is linked to hypertensive heart and kidney disease. Cell. Physiol. Biochem. 15: 109-116.
6. Davis, T.L., et al. 2007. The crystal structures of human Calpains 1 and 9 imply diverse mechanisms of action and auto-inhibition. J. Mol. Biol. 366: 216-229.

CHROMOSOMAL LOCATION

Genetic locus: Capn9 (mouse) mapping to 8 E2.

PRODUCT

Calpain 9 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 Calpain 9 shRNA Plasmid (m): sc-62071-SH and Calpain 9 shRNA (m) Lentiviral Particles: sc-62071-V as alternate gene silencing products.

For independent verification of Calpain 9 (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-62071A, sc-62071B and sc-62071C.

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

Calpain 9 siRNA (m) is recommended for the inhibition of Calpain 9 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

Calpain (B-8): sc-271856 is recommended as a control antibody for monitoring of Calpain 9 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 Calpain 9 gene expression knockdown using RT-PCR Primer: Calpain 9 (m)-PR: sc-62071-PR (20 µl). 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.

PROTOCOLS

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