CRISP-8 siRNA (h): sc-77482



The Power to Question

BACKGROUND

Cysteine-rich secretory proteins (CRISPs) represent a family of evolutionarily conserved proteins which play a role in the innate immune system and are transcriptionally regulated by androgens in several tissues. CRISP-8 (cysteine-rich secretory protein 8), also known as Pl15 (peptidase inhibitor 15), P25Tl or SugarCrisp, is a 258 amino acid secreted protein that belongs to the CRISP family. Expressed at low levels in thyroid, prostate, salivary and mammary tissue, CRISP-8 functions as a serine protease inhibitor that exhibits weak inhibitory action against Trypsin, a serine protease found in the digestive system. In addition to its role as a protease inhibitor, CRISP-8 is secreted in neuroblastoma and glioblastoma cell lines, suggesting a role for CRISP-8 in tumor formation and metastasis within the central nervous system.

REFERENCES

- Giacomoni, D., et al. 1990. Serine proteinase inhibitors produced by human melanoma cell lines. Tumour Biol. 11: 39-50.
- Koshikawa, N., et al. 1996. Purification and identification of a novel and four known serine proteinase inhibitors secreted by human glioblastoma cells. J. Biochem. 119: 334-339.
- Yamakawa, T., et al. 1998. cDNA cloning of a novel trypsin inhibitor with similarity to pathogenesis-related proteins, and its frequent expression in human brain cancer cells. Biochim. Biophys. Acta 1395: 202-208.
- Kaplan, F., et al. 1999. A novel developmentally regulated gene in lung mesenchyme: homology to a tumor-derived trypsin inhibitor. Am. J. Physiol. 276: L1027-L1036.
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CHROMOSOMAL LOCATION

Genetic locus: PI15 (human) mapping to 8q21.11.

PRODUCT

CRISP-8 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CRISP-8 shRNA Plasmid (h): sc-77482-SH and CRISP-8 shRNA (h) Lentiviral Particles: sc-77482-V as alternate gene silencing products.

For independent verification of CRISP-8 (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-77482A, sc-77482B and sc-77482C.

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.

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

CRISP-8 siRNA (h) is recommended for the inhibition of CRISP-8 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 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

CRISP-8 (I-7): sc-130452 is recommended as a control antibody for monitoring of CRISP-8 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 CRISP-8 gene expression knockdown using RT-PCR Primer: CRISP-8 (h)-PR: sc-77482-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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