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PEN-2 siRNA (m): sc-45987

BACKGROUND

Four proteins comprise the γ -secretase complex: Presenilin, nicastrin, Aph-1 and PEN-2. Together, these proteins mediate cell surface signaling pathways for a variety of type I membrane proteins, notably amyloid- β precursor protein, a protein implicated in the development of Alzheimer's disease, via intramembrane proteolysis. The proteins assemble into a proteolytically active complex in the Golgi/*trans*-Golgi network (TGN) compartments. Assembly leads to autocleavage of Presenilin into two subunits to create the active site of γ -secretase, an important step in understanding the mechanisms involved in the etiology and possible treatment of Alzheimer's disease.

REFERENCES

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- Hasegawa, H., et al. 2004. Both the sequence and length of the C terminus of PEN-2 are critical for intermolecular interactions and function of Presenilin complexes. *J. Biol. Chem.* 279: 46455-46463.
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CHROMOSOMAL LOCATION

Genetic locus: Psenen (mouse) mapping to 7 B1.

PRODUCT

PEN-2 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 PEN-2 shRNA Plasmid (m): sc-45987-SH and PEN-2 shRNA (m) Lentiviral Particles: sc-45987-V as alternate gene silencing products.

For independent verification of PEN-2 (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-45987A, sc-45987B and sc-45987C.

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

PEN-2 siRNA (m) is recommended for the inhibition of PEN-2 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

PEN-2 (1C12-G5): sc-293392 is recommended as a control antibody for monitoring of PEN-2 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 PEN-2 gene expression knockdown using RT-PCR Primer: PEN-2 (m)-PR: sc-45987-PR (20 μ l, 438 bp). 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.