SANTA CRUZ BIOTECHNOLOGY, INC.

Endophilin I siRNA (m): sc-35305

BACKGROUND

The Endophilins comprise a family of three SH3 domain-containing proteins designated Endophilin I, II and III, or alternatively known as SH3P4, SH3P8 and SH3P13, respectively. These proteins associate with Amphiphysin, Synaptojanin and Dynamin and are implicated in presynaptic vesicle trafficking at nerve terminals. The expression patterns of the Endophilins are consistent with their cellular functions at the neuronal synapse, as Endophilin I is expressed only in the brain. Both Endophilin II and Endophilin III are detected in a variety of tissues. Endophilin I is also implicated in modulating G protein-coupled receptor signaling by functioning as an adapter protein and directing β1 adrenergic receptors to the endocytic machinery.

REFERENCES

1. Giachino, C., et al. 1997. Novel SH3-containing human gene family preferentially expressed in the central nervous system. Genomics 41: 427-434.
2. Ringstad, N., et al. 1997. The SH3P4/SH3P8/SH3P13 protein family: binding partners for Synaptojanin and Dynamin via a GRB2-like Src homology 3 domain. Proc. Natl. Acad. Sci. USA 94: 8569-8574.
3. Micheva, K.D., et al. 1997. SH3 domain-dependent interactions of Endophilin with Amphiphysin. FEBS Lett. 414: 308-312.
4. Cestra, G., et al. 1999. The SH3 domains of Endophilin and Amphiphysin bind to the proline-rich region of Synaptojanin 1 at distinct sites that display an unconventional binding specificity. J. Biol. Chem. 274: 32001-32007.
5. Schmidt, A., et al. 1999. Endophilin I mediates synaptic vesicle formation by transfer of arachidonate to lysophosphatidic acid. Nature 401: 133-141.
6. Simpson, F., et al. 1999. SH3-domain-containing proteins function at distinct steps in Clathrin-coated vesicle formation. Nat. Cell Biol. 1: 119-124.
7. Tang, Y., et al. 1999. Identification of the Endophilins (SH3P4/P8/P13) as novel binding partners for the β1-adrenergic receptor. Proc. Natl. Acad. Sci. USA 96: 12559-12564.

CHROMOSOMAL LOCATION

Genetic locus: Sh3gl2 (mouse) mapping to 4 C4.

PRODUCT

Endophilin I 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 Endophilin I shRNA Plasmid (m): sc-35305-SH and Endophilin I shRNA (m) Lentiviral Particles: sc-35305-V as alternate gene silencing products.

For independent verification of Endophilin I (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-35305A, sc-35305B and sc-35305C.

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 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

Endophilin I siRNA (m) is recommended for the inhibition of Endophilin I 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

Endophilin I (D-3): sc-48378 is recommended as a control antibody for monitoring of Endophilin I 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 Endophilin I gene expression knockdown using RT-PCR Primer: Endophilin I (m)-PR: sc-35305-PR (20 µl, 600 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.