SANTA CRUZ BIOTECHNOLOGY, INC.

Synaptotagmin XI siRNA (r): sc-270197

BACKGROUND

Synaptotagmins are a large family of synaptic vesicle type III integral membrane proteins that function as regulators of both exocytosis and endocytosis and are involved in neurotransmitter secretion from small secretory vesicles. Synaptotagmins consists of highly conserved C2 domains, which most have a capacity for calcium binding. Calcium-dependent Synaptotagmins act as calcium sensors during vesicular trafficking. Synaptotagmin XI, also known as SYT11 (Synaptotagmin-11), is a 431 amino acid protein that localizes to the membrane and is expressed ubiquitously with highest expression in brain and lung. Unlike other Synaptotagmin proteins, Synaptotagmin XI is suggested to be calcium-independent due to a single point mutation in one of its C2 domains. Synaptotagmin XI interacts with the ubiquitin-E3-ligase Parkin (a juvenile Parkinson's disease gene product), which causes the polyubiquitination and subsequent degradation of Synaptotagmin XI by the proteasome complex. Defects in the gene encoding Synaptotagmin XI are implicated in a number of neurological disorders, including schizophrenia and Parkinson's disease.

REFERENCES

1. von Poser, C., et al. 1997. The evolutionary pressure to inactivate. A subclass of Synaptotagmins with an amino acid substitution that abolishes Ca^{2+} binding. J. Biol. Chem. 272: 14314-14319.
2. Osborne, S.L., et al. 1999. Calcium-dependent oligomerization of Synaptotagmins I and II. Synaptotagmins I and II are localized on the same synaptic vesicle and heterodimerize in the presence of calcium. J. Biol. Chem. 274: 59-66.
3. Mizutani, A., et al. 2000. SYNCRIP, a cytoplasmic counterpart of heterogeneous nuclear ribonucleoprotein R, interacts with ubiquitous Synaptotagmin isoforms. J. Biol. Chem. 275: 9823-9831.
4. Craxton, M. 2001. Genomic analysis of Synaptotagmin genes. Genomics 77: 43-49.
5. Huynh, D.P., et al. 2003. The autosomal recessive juvenile Parkinson disease gene product, Parkin, interacts with and ubiquitinates Synaptotagmin XI. Hum. Mol. Genet. 12: 2587-2597.

CHROMOSOMAL LOCATION

Genetic locus: Syt11 (rat) mapping to 2q34.

PRODUCT

Synaptotagmin XI siRNA (r) 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 Synaptotagmin XI shRNA Plasmid (r): sc-270197-SH and Synaptotagmin XI shRNA (r) Lentiviral Particles: sc-270197-V as alternate gene silencing products.

For independent verification of Synaptotagmin XI (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270197A, sc-270197B and sc-270197C.

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

Synaptotagmin XI siRNA (r) is recommended for the inhibition of Synaptotagmin XI expression in rat 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

Synaptotagmin XI (D-5): sc-365991 is recommended as a control antibody for monitoring of Synaptotagmin XI 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 Synaptotagmin XI gene expression knockdown using RT-PCR Primer: Synaptotagmin XI (r)-PR: sc-270197-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.