SANTA CRUZ BIOTECHNOLOGY, INC.

Munc13-4 siRNA (h): sc-62651

BACKGROUND

Munc13-4 is a member of the Munc13 family and is a homolog to Munc13-1. Munc13-4 lacks the C1 domain and N-terminal extension that are present in other Munc13 family members. It is a peripheral membrane, GTP-Rab 27a-binding protein. Munc13-4 has a ubiquitous tissue distribution; however, unlike related proteins Munc13-1, -2 and -3, Munc13-4 is mainly expressed outside the nervous system. High expression levels of Munc13-4 have been seen in mucous goblet and alveolar type II cells of the lung, as well as in cytotoxic T lymphocytes and mast cells. Munc13-4 localizes to secretory lysosomes. Overexpression of Munc13-4 enhances degranulation of mast cell secretory lysosomes, suggesting that it positively regulates secretory lysosome fusion and exocytosis. Mutations in Munc13-4 cause familial hemophagocytic lymphohistiocytosis subtype 3.

REFERENCES

1. Feldmann, J., et al. 2003. Munc13-4 is essential for cytolytic granules fusion and is mutated in a form of familial hemophagocytic lymphohistiocytosis (FHL-3). Cell 115: 461-473.
2. Shirakawa, R., et al. 2004. Munc13-4 is a GTP-Rab 27-binding protein regulating dense core granule secretion in platelets. J. Biol. Chem. 279: 10730-10737.
3. Neeft, M., et al. 2005. Munc13-4 is an effector of Rab 27a and controls secretion of lysosomes in hematopoietic cells. Mol. Biol. Cell 16: 731-741.
4. Ishii, E., et al. 2005. Genetic subtypes of familial hemophagocytic lymphohistiocytosis: correlations with clinical features and cytotoxic T lymphocyte/natural killer cell functions. Blood 105: 3442-3448.
5. Yamamoto, K., et al. 2005. Mutations of Syntaxin 11 and SNAP 23 genes as causes of familial hemophagocytic lymphohistiocytosis were not found in Japanese people. J. Hum. Genet. 50: 600-603.
6. Hong, W., et al. 2005. Cytotoxic T lymphocyte exocytosis: bring on the SNAREs! Trends Cell Biol. 15: 644-650.
7. Schneider, E.M., et al. 2006. Mutations of perforin and Munc13-4 do not mark HLH by NK defects. Pediatr. Blood Cancer 46: 409-411.

CHROMOSOMAL LOCATION

Genetic locus: UNC13D (human) mapping to 17q25.1.

PRODUCT

Munc13-4 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 Munc13-4 shRNA Plasmid (h): sc-62651-SH and Munc13-4 shRNA (h) Lentiviral Particles: sc-62651-V as alternate gene silencing products.

For independent verification of Munc13-4 (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-62651A, sc-62651B and sc-62651C.

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

Munc13-4 siRNA (h) is recommended for the inhibition of Munc13-4 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

Munc13-4 (C-2): sc-271300 is recommended as a control antibody for monitoring of Munc13-4 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 Munc13-4 gene expression knockdown using RT-PCR Primer: Munc13-4 (h)-PR: sc-62651-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.