

granuphilin siRNA (h): sc-45507

BACKGROUND

Granuphilin, also designated synaptotagmin-like protein 4 or exophilin 2, is a member of the synaptotagmin-like protein family. It is part of a ternary complex consisting of Syntaxin 1A (STX1A) and Rab 27A. The interaction between granuphilin and Syntaxin 1A on the plasma membrane is regulated by Rab 27A. Granuphilin is a peripheral membrane protein mainly expressed in pancreatic β -cells and in the pituitary gland. It has been detected on secretory granules and close to the plasma membrane. In the pancreas, granuphilin interacts with Insulin-containing vesicles while in both the pancreas and pituitary, granuphilin modulates the secretion of hormones and the exocytosis of dense-core granules. Overexpression of granuphilin enhances basal Insulin secretion but also inhibits high K^+ -induced Insulin secretion. The effect of granuphilin on Insulin secretion may be impaired by a mutation that disrupts the binding to either Rab 27A or Syntaxin 1A, making granuphilin a possible regulator in the exocytotic pathway.

REFERENCES

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3. Izumi, T., Gomi, H. and Torii, S. 2005. Functional analysis of Rab 27A effector granuphilin in Insulin exocytosis. *Methods Enzymol.* 403: 216-229.
4. Gomi, H., Mizutani, S., Kasai, K., Itohara, S. and Izumi, T. 2005. Granuphilin molecularly docks Insulin granules to the fusion machinery. *J. Cell Biol.* 171: 99-109.
5. Fukuda, M., Imai, A., Nashida, T. and Shimomura, H. 2005. Slp4-a/granuphilin- α interacts with syntaxin-2/3 in a Munc18-2-dependent manner. *J. Biol. Chem.* 280: 39175-39184.

CHROMOSOMAL LOCATION

Genetic locus: SYTL4 (human) mapping to Xq22.1.

PRODUCT

granuphilin 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see granuphilin shRNA Plasmid (h): sc-45507-SH and granuphilin shRNA (h) Lentiviral Particles: sc-45507-V as alternate gene silencing products.

For independent verification of granuphilin (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-45507A, sc-45507B and sc-45507C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

granuphilin siRNA (h) is recommended for the inhibition of granuphilin 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 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

granuphilin (C-3): sc-374544 is recommended as a control antibody for monitoring of granuphilin 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 granuphilin gene expression knockdown using RT-PCR Primer: granuphilin (h)-PR: sc-45507-PR (20 μ l). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.