SANTA CRUZ BIOTECHNOLOGY, INC.

Rabenosyn-5 siRNA (m): sc-76334

BACKGROUND

Zinc-finger proteins contain DNA-binding domains and have a wide variety of functions, most of which encompass some form of transcriptional activation or repression. Rabenosyn-5, also known as ZFYVE20 (zinc finger, FYVE domain containing 20), is a 784 amino acid cell membrane protein that contains a C_2H_2-type zinc finger, a FYVE-type zinc finger and a ubiquitin-interacting motif (UIM repeat). The FYVE domain is a cysteine-rich domain of about 70 amino acids. Its primary role is to target signal-transducing proteins to cell membranes through binding to the membrane lipid PIP3 (phosphatidylinositol-3-phosphate) with high specificity. Considered an effector protein, Rabenosyn-5 is required for endosome fusion either homotypically or with clathrin coated vesicles and is involved in the lysosomal trafficking of cathepsin D from the Golgi to lysosomes. Rabenosyn-5 promotes the recycling of transferrin directly from early endosomes to the plasma membrane and binds phospholipid vesicles containing PIP3.

REFERENCES

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2. Hu, Y., et al. 2002. SARA, a FYVE domain protein, affects Rab 5-mediated endocytosis. J. Cell Sci. 115: 4755-4763.
3. de Renzis, S., et al. 2002. Divalent Rab effectors regulate the sub-compartmental organization and sorting of early endosomes. Nat. Cell Biol. 4: 124-133.
4. Naslavsky, N., et al. 2004. Rabenosyn-5 and EHD1 interact and sequentially regulate protein recycling to the plasma membrane. Mol. Biol. Cell 15: 2410-2422.
5. Eathiraj, S., et al. 2005. Structural basis of family-wide Rab GTPase recognition by Rabenosyn-5. Nature 436: 415-419.
6. Gengyo-Ando, K., et al. 2007. The SM protein VPS-45 is required for RAB-5-dependent endocytic transport in *Caenorhabditis elegans*. EMBO Rep. 8: 152-157.
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8. Fichtman, B., et al. 2008. EHDS are serine phosphoproteins: EHD1 phosphorylation is enhanced by serum stimulation. Cell. Mol. Biol. Lett. 13: 632-648.
9. Naslavsky, N., et al. 2009. EHD3 regulates early-endosome-to-Golgi transport and preserves Golgi morphology. J. Cell Sci. 122: 389-400.

CHROMOSOMAL LOCATION

Genetic locus: Zfyve20 (mouse) mapping to 6 D1.

PROTOCOLS

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

PRODUCT

Rabenosyn-5 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 Rabenosyn-5 shRNA Plasmid (m): sc-76334-SH and Rabenosyn-5 shRNA (m) Lentiviral Particles: sc-76334-V as alternate gene silencing products.

For independent verification of Rabenosyn-5 (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-76334A, sc-76334B and sc-76334C.

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

Rabenosyn-5 siRNA (m) is recommended for the inhibition of Rabenosyn-5 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Rabenosyn-5 gene expression knockdown using RT-PCR Primer: Rabenosyn-5 (m)-PR: sc-76334-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.